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PHYTOHORMONES (ORGANIC ACIDS) IN RELATION TO THE ROOTING OF CERTAIN HORTICULTURAL PLANTS.

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APRIL, 1939.

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Charles Raymond Ure,
Department of Horticulture.

A Thesis

submitted to the University of Alberta
to fulfil approximately one-half of the
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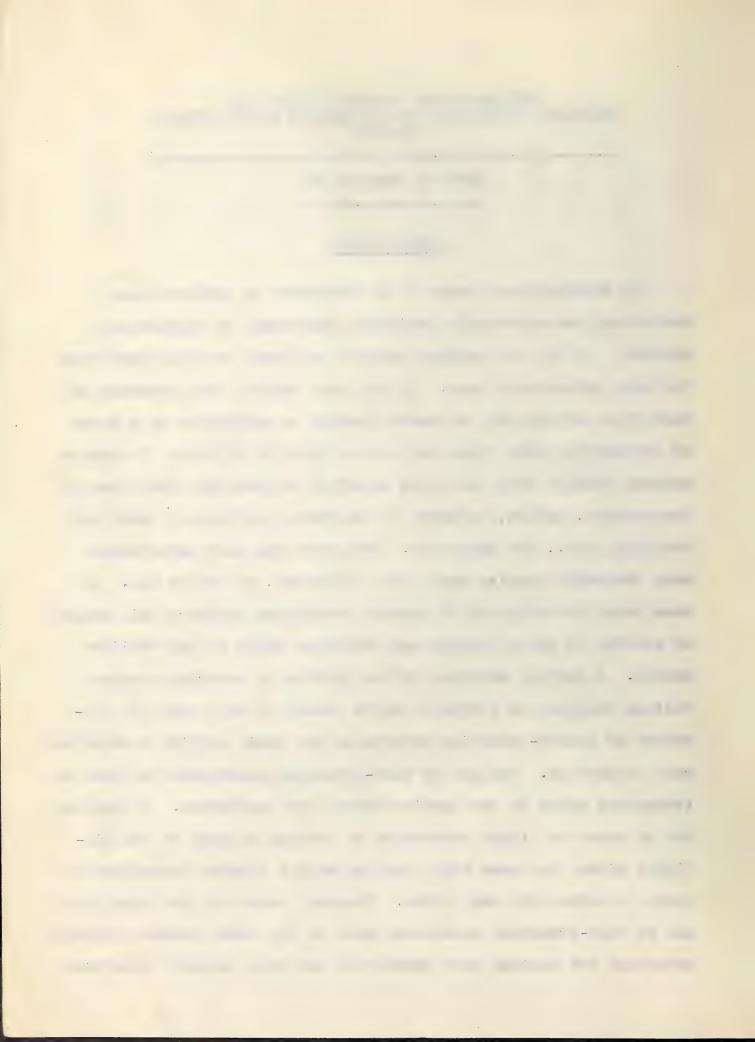
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PHYTOHORMONES (ORGANIC ACIDS) IN RELATION TO THE ROOTING OF CERTAIN HORTICULTURAL PLANTS.

Charles Raymond Ure

INTRODUCTION

In horticultural crops it is necessary to increase many varieties, and especially desirable specimens, by vegetative methods. Of all the asexual methods employed, cuttings have been the most extensively used. In the last decade, the greenwood or half-ripe cutting has increased greatly in popularity as a means of propagating many trees and shrubs grown in Alberta. To ensure maximum results with this type carefully controlled conditions of temperature, medium, maturity of the wood, position of basal cut. humidity, etc., are necessary. Even with the best environment many desirable species root with difficulty, or not at all. some cases the adoption of special conditions induce a fair amount of rooting in those species and varieties which do not "strike" easily. A partial solution of the problem of securing adequate rooting response in difficult sorts seemed at hand with the discovery of growth-promoting substances and their ability to stimulate root production. The use of root-promoting substances has been of tremendous value to the horticulturist and nurseryman. It enables him to obtain a higher percentage of rooting in many of the difficult kinds, and more rapid rooting with a greater production of roots in moderately easy kinds. However, even with the beneficial use of root-promoting substances most of the other factors formerly necessary for maximum root production are still equally important.

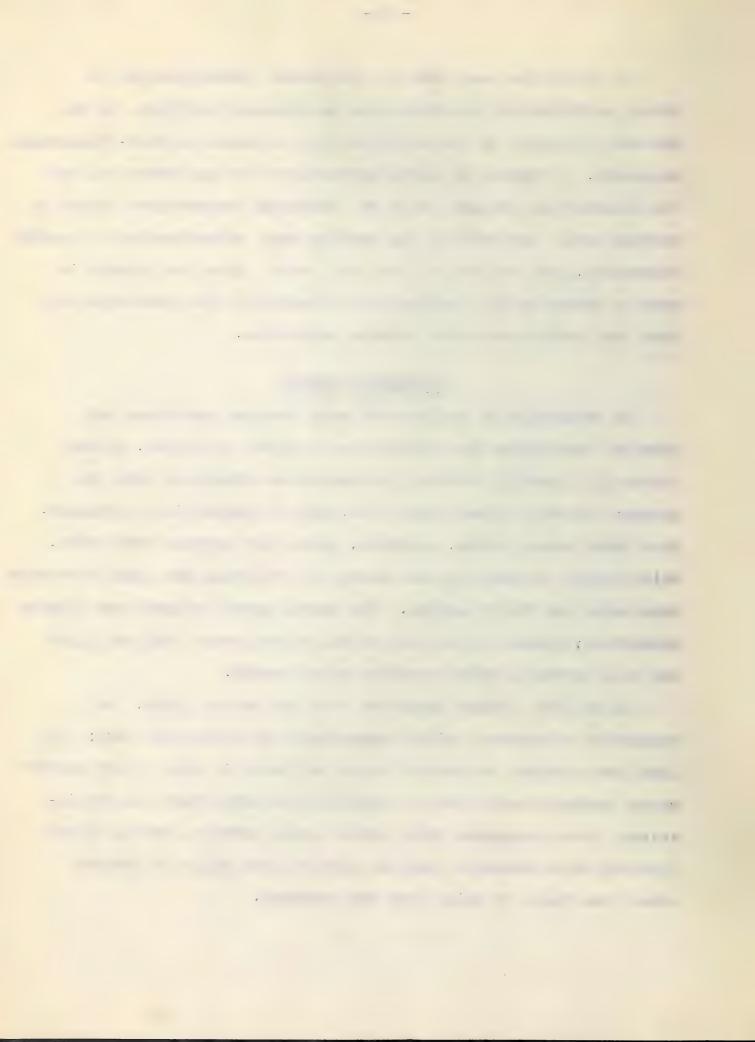


An effort has been made in the present investigations to obtain satisfactory root formation on greenwood cuttings, in ten species of plants, by the application of a number of root-stimulating compounds. Attention is given particularly to the effect of, and the interactions brought about by, different temperatures, types of rooting media, maturity of the cutting wood, concentration of growth-substances, and position of the basal cuts. Also, an attempt is made to determine the combination of variables that encourage maximum root production under Alberta conditions.

LITERATURE REVIEW

In attempting to analyze the many chemical reactions and physical conditions that contribute to growth in plants, Boysen Jensen (2), usefully divides the substances concerned into two groups: (a) nutritional substances, and (b) regulating substances. In a broad sense, water, minerals, gases, and organic foodstuffs, which supply the material and energy for building the plant structure come under the first heading. The second group is much less clearly understood; indeed, it is only within recent years that any light has been thrown on this aspect of plant growth.

About 1935, Huxley suggested that the second group, the regulating substances, might conveniently be subdivided into: (1) localized chemical activators whose influence is over a very narrow range, perhaps restricted to intracellular activities, as for instance, those concerned with various genic effects, and (2) plant hormones which exercise specific effects upon cells or tissues other than those by which they are produced.



Explanation of Terms.

Unfortunately, many terms have been used interchangeably and often rather loosely in the literature. A clear distinction has not always been maintained between substances which may be correctly termed hormones and certain other types of materials. The word hormone is of Greek origin meaning "I arouse to activity," was suggested by Hardy, and first applied in animal physiology by Starling in 1906. He defined it as "any substance normally produced in the cells of some part of the body and carried to other parts which it affects for the good of the body as a whole." Hormones are chemical substances which have a specific influence on correlation and differentiation of the organism. For many years they were thought to have occurred in animals only and had not been associated with growth in plants until quite recently, even though Fitting in 1910 used the word hormone in connection with plants, when he noticed that a substance present in orchid pollen caused a swelling of the gynostemium of the orchid flower. Hormones are effective when present in very minute quantities and control growth in plants by some means other than by direct nutrition. In animals there are substances of accessory or protective food nature which do influence growth through direct nutritive means. These have been classed as vitamins. In certain plants, Boysen Jensen (1), (2), Miller (22), and Kögl (12), have found substances that apparently function much like vitamins and have been termed bios. Besides vitamins and bios, another class of substances, enzymes, are produced by living organisms which promote chemical reations. These three are what has been referred to above as nutritional substances.

Plant hormones have been variously referred to as growth hor-

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mones, growth regulators, growth enzymes, phytohormones and auxins. Many chemical growth-promoting substances have been synthetically prepared and used by Hitchcock and Zimmerman (40), Manske and Leith (20), and others, and it seems advisable to differentiate between these substances and those spoken of as hormones. Growth-promoting substances are synthetic chemical compounds which induce hormonelike responses in plants. Most of them stimulate root development. Since they produce such responses, it seems reasonable to assume that some of them at least may be found in living plant tissues. However, up to the present none of these chemicals (some fifty in number) worked on by Hitchcock and Zimmerman (40) have been isolated from green tissue. Whether or not they will be found in green plants is of course unknown. For the present, at least, it seems necessary to distinguish between the synthetic substance and those found associated with living tissues (hormones). The term phytohormone was introduced to designate hormone-like substances in plants, to avoid confusion with the animal hormones, but it is now widely used to refer to the synthetically prepared growth-promoting substances. Throughout the remainder of the text the term phytohormone is used to designate this group of synthetic, growthpromoting substances.

Some workers use the term auxin to indicate this heterogeneous group of substances which bring about specific growth reactions.as distinguished from those substances causing cell elongation, and whose activity is measurable on higher plants. In addition to the synthetic, stimulative compounds, another group extracted from animal and lower plant materials are included in the larger group known as auxins. The three main growth-substances from animal and plant sources are hetero-auxin, and auxin a and b. Contrary to

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reports heard about hetero-auxin (indole-3-acetic acid), or auxin a and b, so far they have not been extracted from green tissues of higher plants. Some fungi have been reported to produce hetero-auxin, and auxin a and b, but the part these three actually play in the growth of green plants is not definitely known.

Theories in Regard to Regeneration.

From the earliest times men have tried to explain why isolated pieces, stems or leaves of many plants had regenerated and become independent individuals. They wondered what the factors were which set up this regeneration, and which determined the kinds of organs produced and the manner of their formation. By the latter part of the 19th century the phenomenon of correlation between the many plant functions was occupying the attention of men. One of the first attempts to explain regeneration was by supposing the existence of "Anlagen" (Goebel). Anlagen is apparently derived from Anlage meaning the first trace of an organ. Malphighi, in 1675, and Agricola, in 1716, had expressed similar vague ideas on this correlation phenomenon.

Two-sap Theory.

About 1758, experiments by Duhamel du Monceau led him to believe correlation was broughtabout by two saps, one moving upward, the other downward, from the leaves, through the cortex to the roots. It was supposed that if this downward stream was intercepted in any way the sap caused callus formation from which roots arose. So much stress was laid on root formation that the swelling and callus at the point of interception were considered as being much the nature of roots.

Special Substance Theory.

For the next hundred years the physiological concept of

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correlation seems to have been forgotten. By about 1860 began the rapid development of plant physiology and the correlation phenomenon again came into prominence. Careful studies by Sacks, reported in 1880, 1882 and 1893, brought forward the "complete" theory, which is sometimes referred to as the "special substance" theory. This was really a modern view of Duhamel's conception. Sacks believed that the morphological differences between plant organs was due to corresponding differences in their material composition, which must be present at the time of their initiation, even though at that stage of development chemical reactions or other crude methods failed to show any differences. To account for these differences he assumed special substances governing root-formation, flowerformation, etc., which move in different directions in the plant. Root forming substances were believed to form in the leaves and being heavy moved downwards to the base. These substances he termed "Wuchsstoffe."

Nutritional-basis Theory.

In the years between Duhamel's "two-sap hypothesis" and the "special substance theory" of Sachs great advances were made in the knowledge of plant morphology and the inherent nature of the tissues themselves. These studies led to the belief that differences in plant organs were due to nutritional differences. Following Sachs most of the studies on correlation placed the emphasis on nutritive factors. Goebels in 1908 rejected the special substance idea in favor of a nutritional basis. He pointed out that numerous organ initials may remain undeveloped because the building materials, which they need for their development, go to others which can attract these materials more strongly. Similar views have been held by other workers, Kraus and Kraybill (13), who noted the

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relationship between a proper balance of carbohydrate, nitrogen and moisture with vegetation and fruitfulness in the tomato.

However, it was Loeb (19) who, experimenting on the regeneration of Bryophyllum calycium, observed that a stem segment bearing leaves will form roots more readily than one from which the leaves are removed. His observations led him to again stress the theory of hormones and to suggest that they play a great part in root formation. Van der Lek (15, 16) also found that the presence of leaves or buds promote the formation of roots at the basal end of cuttings.

Tropisms.

Before this time, about 1880, tropisms were being recognized as a special kind of correlation phenomenon. The intriguing subject of tropisms has received much attention and resulted in many of the newer concepts of growth. The aspect of tropisms was particularly emphasized by Darwin (6). From his invaluable experiments on the movement of plants in response to light he concluded there must be present some substance in the upper part of the plant which is acted upon by light and which transmits its effect to the lower parts, for example, causing a bending when unilateral light is supplied. Rothert's work, about 1894, completely substantiated Darwin's findings. Fitting, from 1905 to 1907, carried Rother's work further and by making incisions in the Avena coleoptile before applying light concluded that the stimulus responsible for curvature is conducted around the incision and transmitted exclusively through the living material. Thus the conclusions which have been drawn from Fitting's experiments, which is essentially Darwin's conception, is that when seedlings are freely exposed to a lateral light, some influence is transmitted from the upper to the lower part causing the latter to bend.

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Boysen Jensen from 1910 to 1913 was able to show that a phototropic stimulus can be transmitted across a wound gap where the illuminated tip of Avena coleoptile is connected to the lower portion by means of a strip of gelatin, and concluded that the stimulus is of a material nature. Paål in 1914 and 1919 repeated and extended Boysen Jensen's experiments taking care to eliminate any possibility of the stimulation coming in from any source other than the illuminated tip. His extensive experiments made it evident that the tip is the seat of a growth regulating center.

Thus Paal was able to establish the theory that growth of the coleoptile is controlled from the tip by means of a diffusable substance. This was later confirmed by careful growth measurements of Söding in 1923 and 1925. Paal's view was further substantiated by Cholodny and by Went who attributed all tropisms to asymmetric distribution of the normal growth-promoting substance.

Results of experiments on five species of plants - tomato, sunflowers, cosmos, marigold and tobacco - by Zimmerman and Hitchcock (38), involving the treatment of intact plants, plants with tops removed or excised shoots, with phytohormones show that induced tropisms are conditioned by the relative positions of the organs to gravity. For example, horizontal stems which normally bend away from the earth were induced to curve towards the earth by applying certain concentrations of water solutions of phytohormones to the basal end or tip of the shoot. With the same concentrations leaves of upright shoots showed epinasty whereas leaves of inverted shoots showed hyponasty. These and somewhat similar results give some evidence that synthetic growth substances, like natural hormones, are unequally distributed through plant tissues.

It was well established by this time that growth substances or growth hormones are functional in correlation and growth. The

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realization of this fact gave added impetus to research along this line and since about 1925 on, literature on the subject has become voluminous.

Isolation and Sources of Growth Hormones.

The presence of growth hormones being proven and their functional relation to growth established, the next research step was to isolate them and attempt to produce the typical response by applying the hormone externally. First attempts to extract growth substances with agar blocks were unsuccessful and it was not until about 1928 that Went finally obtained the active substance from the coleoptile tip. By improvement in technique he was able to demonstrate in the Avena coleoptile the presence of an active substance from the tip by extraction with agar. This was the final convincing step that proved beyond a doubt the presence of plant hormones. Went's experiments led to what is known as the "Avena test", which is now used extensively in detecting the presence of growth hormones. Though it has been unquestionably demonstrated that growth substances do occur in higher plants, they are present in such minute quantities that no satisfactory means of microchemical detection has been devised as yet.

Synthetic Preparation of Growth-Substances.

Growth substances have been prepared from plant and animal material of many sources. These chemically pure substances have been collectively termed auxins, which is often used interchangeably with the physiological terms growth substances, growth hormones, etc.. From fungus cultures of such species as Rhizopus suinus, and Asperigillus niger and Yeast has been prepared the growth substance hetero-auxin (3-indole-acetic acid). This compound has also been obtained from urine and prepared by synthetic

methods. Besides hetero-auxin, Kögl (12) and others, isolated from human urine another active substance which was termed auxin a.

Auxin a or auxentriclic acid has the formula $C_{18H32}O_{5}$. Another substance which appears to be an isomer of auxin a has been obtained from malt and corn germ oil and has been named auxin b. Auxin b or auxenolonic acid has the formula $C_{18H30}O_{4}$. The activity of indole-3-acetic acid derived from urine or fungal growth is of the same order as the C_{18} compounds auxin a and b and is not due to any impurities since the pure synthetic compound has the same activity. As Went (31, 32) has pointed out these three auxins are physiologically indistinguishable, as all of them give the same type of growth response.

Following the discovery of the activity of indole-3-acetic acid, a large number of synthetic substances which are more or less active as growth substances, have been discovered and recognized by Zimmerman and Wilcoxon (41), Zimmerman, Hitchcock and Wilcoxon (40), and Manske and Leitch (20). Thimann (27, 28) proceeded to physiologically analyze the differences in activity by comparing the three substances: (1) indole-3-acetic acid, (2) indene-3-acetic acid, (3) cumaryl-l-acetic acid. He found the latter substance to have no activity in producing curvature in Avena, but did possess moderate activity in straight forward growth of Avena sections and still more activity in producing pea stem curvature. Indene-3-acetic acid produced similar results, though less extreme. Thus, a substance may be active in growth promotion without causing curvature on Avena. These substances which cause relatively little or no curvature on Avena are regarded as possessing primary growth promoting activities. Further tests, by Went and Thimann (33), of many growth substances indicated that primary growth-promoting

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activity is connected with the presence of: (1) the double bond, or aromatic saturation; (2) a carboxyl group, free, or if esterified, readily hydrolizable; (3) a ring system, either 5 membered (auxin a and b), aromatic (naphthyl or phenyl), or a combination of both (indole, indene, etc.); (4) a minimum distance of at least one C atom between the carboxyl group and the ring; (5) a very definite steric structure. Other properties of the growth substances, such as length of the acid side chain or the structure of the nucleus itself, have been found to modify the secondary properties of a growth-promoting substance. These do not directly affect the primary growth reactions. Such facts have made prediction of substances that should show activity quite accurate.

Hormones in Relation to Root Initiation.

Up to the present, reference has been made largely to the relation of auxin to shoot growth. In roots, the relationships are somewhat different, and yet, in some respects similar. The influence of many kinds of substances upon the initiation and growth of roots has been studied by numberous workers, but only within recent years has it become apparent that Sach's hypothesis in 1882 of root-forming substances may have some actual basis. As previously stated, decapitation of the shoot invariably results in reduction of growth rate. Decapitation of the roots, on the other hand, does not greatly affect growth. This point was much investigated around 1870-80, but probably due to many conflicting results the whole question was dropped for nearly 50 years.

As previously pointed out, in early attempts to explain root formation Duhamel, Sachs, and others, assumed the accumulation of special root-forming substances at the base of the cutting. This was later followed by attempts to correlate root formation and

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growth with nutritive factors. However, Loeb's experiments (17, 18, 19) showed that stem segments of Bryophyllum (house leek family) form roots and bend geotropically more readily when leaves are attached to the stem. From this fact Loeb says "All these facts suggest a close association if not identity between the root forming substances and the substances or hormones causing geotropic curvature."

The conclusions of Loeb were corraborated by Van der Lek in 1925, and this led to further investigation as to the possible role of hormones in root initiation. Van der Lek distinguished between roots which develop from preexisting root initials and those which are newly formed after treatment. Went pursued this new line of attack and was able to extract from leaves and germinating barley a non-specific, heat-resisting substance that caused the initiation of roots when applied to cuttings. The function of root-forming substances was further studied in such species as Ribes, Salix, Populus, and Vitis by Van der Lek (15, 16), who observed that buds, especially rapidly developing buds, promoted strong root formation, but if the buds were removed root formation practically ceased unless the species possessed already developed root primordia. It was also noted that completely dormant poplar cuttings taken in winter no longer promoted root development, and may even inhibit it until the dormancy is broken. Leaves were less effective in promoting root formation than strongly developing buds. Similar results were found in Impatiens and Acalypha by Went and others. It is quite evident that root-formation is due to a special substance or hormone, which was termed rhizocaline by Went. Rhizocaline was believed not to be a nutrient, was found to be thermo-stable, and was produced by leaves in light. It was also stored up in

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cotyledons and buds, and its transport was basipetally polar.

Following the discovery and isolation of auxin a and b and indole-3-acitic acid, and the fact that they were extremely active in root formation, Thimann and Koepfli (29), Kögl (12) demonstrated the similarity between these substances and rhizocaline and the latter name was dropped. However, not always was root forming and growth promoting activities of the natural preparations (auxins) quantitatively parallel to the root-forming substances. The differences were later found to be due to such secondary factors as carbohydrates, bios, and interrelations between auxins.

The formation of roots on cuttings as well as on intact plants by the application of auxins and synthetic growth promoting substances has been studied by many investigators, Went and Thimann (33), nurserymen and others. Their researches demonstrate that growth substances, both natural and synthetic, not only hasten the rooting of cuttings which ordinarily root, but stimulate many plants which would not otherwise root and also increase the number of roots on those that normally produce only a meagre supply. Thus, it appears that those substances causing cell elongation may also be effective in root formation.

Zimmerman and Hitchcock (39) and Zimmerman, Crocker, and Hitchcock (35, 36), have shown that auxins are not alone in their ability to produce new roots and have demonstrated the initiation and stimulation of adventitious roots by treatment with appropriate doses of acetylene, ethylene, propylene, and carbon dioxide on some 15 species and varieties of plants. They concluded that ethylene may act as a hormone; but it has since been shown that the effect of ethylene is to inhibit and not to promote elongation. Experiments by Michener (21) seem to indicate that ethylene is

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explains the results obtained by Zimmerman, Crocker, and Hitchcock (35) since their experiments were carried out on green plant in light, and were likely well supplied with auxin. The role of light in root formation is not well understood as the results of investigators have been very conflicting. It has been shown in some species which are difficult to root, e.g. apple, that when the twigs are etiolated for some time before the cuttings are taken they root much better. On the other hand, it is well known that leafy cuttings require light in order to root. Results of experiments by Went and Thimann (33), indicate that cuttings having a storage of auxin, such as dormant deciduous plants, will root best in darkness, whereas leafy cuttings without a storage of auxin require light for auxin formation which is necessary for rooting. Zimmerman and Hitchcock (37) corroborate these results.

Manske and Leitch (20), Zimmerman and Hitchcock (41), Zimmerman et al (40), have prepared and tested many different growth substances and have compared the effectiveness in initiating roots on stems, leaves and cuttings when applied either in the form of lanoline paste or in solution. Their studies indicated that B-indole-butyric acid, B-indole-acetic acid, and alpha-naphthaleneacetic acid are the most effective substances for the stimulation of new roots.

The practical application of auxins and phytohormones in rooting cuttings of commercially important plants was begun by Cooper (4). Working with Citrus, Acalypha, Lantana, and Tradescantia, he obtained excellent root formation, using pure synthetic B-indolyl-acetic acid applied to the cuttings by the lanolin method of Laibach. He concluded that the hormone not only increase rooting

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on cuttings which will root when untreated, but also cause the formation of roots on leafless <u>Citrus</u> and <u>Tradescantia</u> cuttings which do not ordinarily form roots. In a succeeding experiment Cooper (5), noted that the solution method of treatment is more effective on rose, lemon, holly and chrysanthemum cuttings than the landlin method. This fact has since been further substantiated by numerous other workers. Though Cooper (5) observed that basal treatment is more effective than application of the phytohormone to the tip, this is just a verification of an important fact noted earlier.

Most of the recent studies in the practical utilization of these compounds have been directed towards their use as aids in the rooting of cuttings. Grace (7) in an extensive investigation studied the relation of some of these compounds to seeds, growing plants, cuttings, and lower plant forms. His preliminary results in the case of seeds of wheat and barley showed an increase of 102 per cent in root length when the seeds were previously treated with a dust consisting of a mixture of 2 p.p.m. of indole-acetic acid and 2 p.p.m. of naphthylene-acetic acid plus standard mercurial disinfectant dust. Overdosages depressed both root and tip growth. Top growth of plants from treated seed was claimed to have been increased 20 to 30 per cent over the controls. He also found that in the case of growing plants the correct range of dosage gives a very definite and clear cut stimulation to root and top growth, and that very young plants react more favorably than do older ones. The result in the case of the cuttings was just as favorable as has been reported by other workers. This experiment indicated that further practical use may be made of the stimulative action of phytohormones. Other reports of experiments along this line have

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not been so encouraging. Proper concentration is an extremely important factor. The relation of different concentrations of auxin to root, bud and stem growth has been shown by Thimann (25). He showed that proper concentration will stimulate root, bud or stem growth whereas overdosages inhibit growth.

However, the question of root stimulation on cuttings is not as simple a matter as it might first appear. Many factors enter into the situation, such as temperature, moisture, oxygen supply and species reaction, and root formation is the net result of the interaction of all necessary components. The role of sugars, light, water, oxygen, auxins, and sensitivity of the species is a good example of an interlocking system of limiting factors. Root formation may vary depending upon the variation from the optimum of any one of these factors; and when any one of the above factors is lacking altogether root production will not occur. As already indicated many synthetic growth substances are effective in the production of visible roots. The mechanism of action of these different compounds is not known. They may act directly, or through some influence upon the auxins, or perhaps in still other ways.

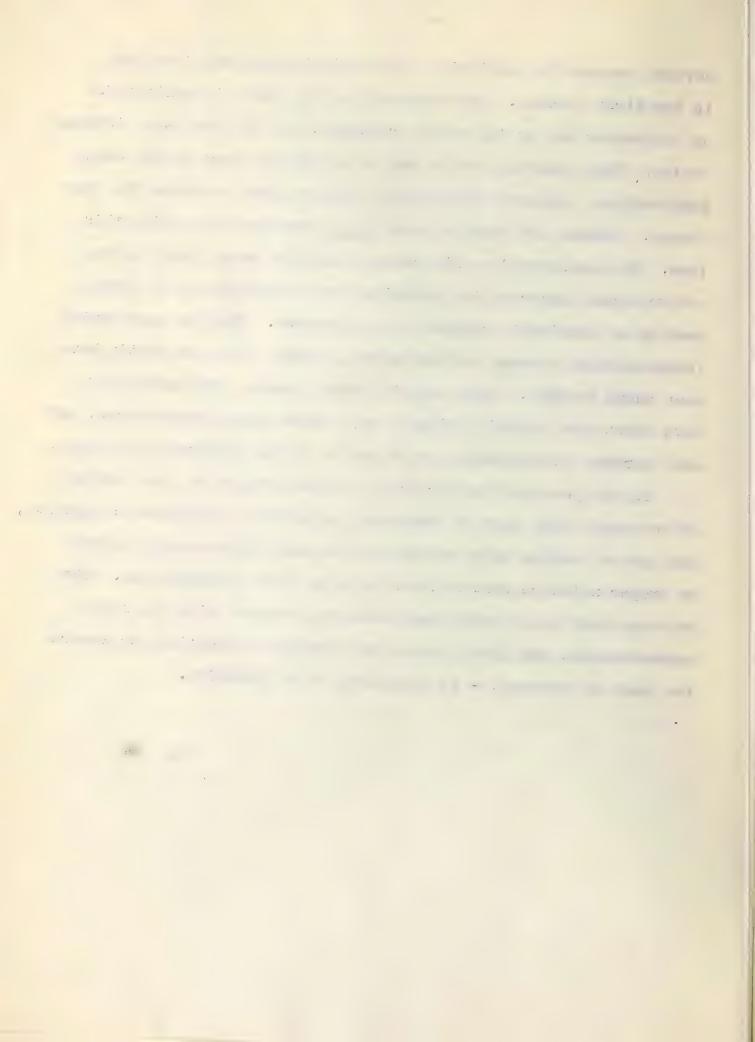
Thus far, our consideration has been with those factors which cause the production of visible roots. Here again the stage of development at which each factor exerts its influence is unknown. In order to produce a visible root three processes must take place in succession. There must be (a) the redifferentiation of the pericycle cells into a root initial, (b) the development of the initial into a root primordium, and (c) the root primordium must develop into a root by outgrowth. The first two may be regarded as one process as the changes may have to take place after the chemical treatment or they may have occurred before the cutting was taken. Thimann and Went (91) pointed out that there are

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several reasons for believing that auxin in some way functions in the first process. The outgrowth of the roots is believed to be influenced not by the auxin treatment, but by some other internal factor. This internal factor may be attracted there by the auxin application. Cooper's experiments (5) give good evidence for this theory. Pearse (23) working with Salix does not agree with this idea. He concludes from his results that the acid itself is the active agent concerned in promoting root formation and is itself used up or chemically changed in the process. Similar complicated interrelations between carbohydrates, auxins, bios and biotin have been shown by Kögl. More recently Went, Bonner, and Warner (34) have shown that aneurin (vitamin B1), under some circumstances, may act, perhaps in conjunction with auxin, in the production of roots.

In the practical application of phytohormones to the rooting of cuttings other factors including temperature, atmospheric humidity, and type of medium which allow for adequate oxygen supply as well as proper moisture content must be given some consideration. The cuttings must be of sufficient maturity, treated with the right concentration, and given proper environmental conditions to secure the desired response, - in this case, root formation.



PRELIMINARY STUDIES.

During the summers of 1935 and 1936 preliminary tests were conducted on the rooting of certain Alberta grown trees and shrubs, by means of softwood cuttings. The softwood or greenwood cutting is a cutting taken from woody or herbaceous plants in the summer months, usually in June, July or August, from growing, immature, leafy shoots. The rooting response, of this type of cutting, in relation to such factors as the proper time to take the cuttings, best type of medium, optimum temperature, most suitable place of the basal cut, and determination of the kinds which root most readily, were some of the problems given consideration.

It was felt that if satisfactory rooting could be obtained it would be of considerable benefit to western nurserymen; for up to the present time, in Alberta, nurserymen practise propagating only a few kinds of outdoor plants, by means of stem cuttings. The custom has been to import lining-out material, usually from Holland. Unfortunately, this practice has not always given the most satisfactory or economical results.

A few of the general conclusions from these tests, reported by Ure (30), will be cited as they form a basis upon which the work reported herein was planned.

"These preliminary tests demonstrate that many Alberta grown trees and shrubs can be propagated by means of greenwood cuttings. In most of the shrubs tested, some 30 kinds in all, the stage of maturity, which ensured good rooting, covers a sufficient period of time to enable extensive use of this method of propagation. It is likely that wider application of softwood cuttings could be made use of by nurserymen in the west, with both profit to themselves and the general public."

The purpose of the preliminary tests was to secure some

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indication of the rooting response under Alberta conditions, of as many species as possible. Some species of plants were found to root readily in three to four weeks, whereas others produced no roots even after eight to ten weeks in the rooting medium.

Many of the sorts which root with difficulty, or not at all, are desirable kinds and it was decided to investigate further the possibilities of inducing roots by use of plant-growth substances. With this object in view, and keeping in mind also, the aforementioned factors, namely, type of medium, temperature, response of different species, and place of the basal cut, the present work was undertaken.

1937 EXPERIMENT.

Materials.

Plant Materials.

The plant materials, used in the 1937 experiment, were carefully selected as to type, maturity and uniformity. As in the preliminary tests, only greenwood cuttings were used. It was found necessary to eliminate many desirable species, because of the large amount of material required, and the lack of sufficiently uniform cutting wood. All the cuttings collected were from species growing on the University grounds. Four species were tried, namely:

Amelanchier alnifolia, Cotoneaster acutifolia, Lonicera tatarica, and Syringa villosa.

Media.

In the preliminary tests previously referred to, three types of media were used. These were sand, peat moss, and a mixture of sand-and-peat moss, in equal parts by volume. Indications, from this earlier work, were that peat does not give as satisfactory

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results as do the other two. For this reason, and because it was necessary to reduce the number of variables, so that more important aspects of the rooting problem might be adequately tested, only two types of medium were used in the 1937 work. The two media consisted of sand and the sand-and-peat mixture. The sand was a clean. reasonably fine grade obtained from dealers in Edmonton. The peat moss was a partly decomposed and finely ground product obtained from peat bogs west of the city. The media, before being used. were sterilized, in an autoclave at 30 pounds steam pressure, for 10 to 12 hours, to kill soil-borne disease organisms. Also, the frames were thoroughly sterilized with a strong formaldehyde solution, 1 pint to 30 gallons of water, the day prior to placing the media in them. For the sand-and-peat mixture, the ingredients, in equal parts by volume, were well mixed before placing in the rooting-frame. Before planting the cuttings, pH determinations of the sterilized media showed the sand to be slightly alkaline (pH 7.5), and the mixture to be nearly neutral (pH 6.8).

Phytohormone Material.

Pure, synthetic, organic acids were used as the root-promoting substances. Four acids were tested in 1937 consisting of indoleacetic. indole-butyric, indole-propionic and naphthalene-acetic. These materials, obtained from Dr. R. H. Manske of the National Research Council, Ottawa, Canada, were all in crystalline form, but were used in solution as outlined later.

Frames.

The cuttings were rooted in a series of frames in the greenhouse. These frames. 31 feet wide by 6 feet long, were constructed of 12 inch cedar, and equipped with close fitting sash. Four such frames were placed end-to-end on a bench in the greenhouse, Fig. 1.

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Fig. 1. An external view of the rooting frames showing the arrangement in the greenhouse.



Fig. II. Inside view of one of the rooting frames.



An inside view of one of the frames may be obtained from Fig. 2.

Overhead, a light grade of white canvass cloth was arranged so as
to provide shade when necessary. Two of the four frames were
equipped with thermostats in order to provide for bottom heat.

Method.

Outline of Experiment.

Only one extensive experiment was conducted during the summer of 1937. In this work, the following factors were given consideration; type of medium, temperature, species, phytohormone treatment, concentrations of the growth-substances, and place of the basal cut.

Media.

Two types of rooting media were employed, which consisted of sand and a mixture of sand-and-peat. These were treated and prepared as already outlined under "Materials."

Temperature.

The effect of two temperatures was studied. In the preliminary tests of 1935-36 three temperature conditions were tried, namely; one at uncontrolled greenhouse temperature; the other two were thermostatically controlled at 70° F. and 80° F., respectively. The former temperature fluctuated to some extent with changes in outside atmospheric conditions. The preliminary work indicated that most species do not root satisfactorily at 80° F. In order that space might be available to test more important factors, the 80° F. temperature was omitted from the 1937 plan. The two temperatures used were; 70° F. and the uncontrolled greenhouse condition, which served as a check. A record of the temperature within the frames was kept. The average temperature in the check frame, over the period of the experiment, was found to be slightly over 60° F.

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This was somewhat lower than the greenhouse temperature, due to the fact that the temperature in and around the frames was kept reasonably constant by shading and frequent sprinkling of the frames and walks with water on hot days.

Phytohormones.

The four organic acids employed were applied in the form of aqueous solutions to the base of the cuttings. The proper concentration of phytohormone is an extremely important factor in securing good rooting response. Limited rooting space made it impossible to test many concentrations adequately, and still study the effect of the other variables. To offset this disadvantage, three rather widely differing concentrations were used with the hope that they would give some indication of the limits for each species. Subsequent work would then be necessary to determine the optimum concentration for each acid. The three concentrations used consisted of: (1) 0 concentration, which served as a check; (2) 20 mg. per 100 cc. of water; and (3) 40 mg. per 100 cc. of water. Basal Cut.

It was thought advisable also to study the effect of the place of the basal cut. The common practice, in the propagation of trees and shrubs, is to recommend making the basal cut at one of three places; (a) at the node, (b) one-half inch below the node, or (c) one-half inch above. In the present work the effect of the first two recommended cuts, namely, at the node, and below, were studied, in their relation to rooting response.

Design of the Experiment.

The entire experiment was so planned that statistical analysis might be applied to the results. The experiment was done in duplicate, and was designed to study the effect of the following

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factors in root initiation:

- 1. Two types of media. -
 - (a) Sand. (S)*
 - (b) Sand-and-peat. (S+P).
- 2. Two temperatures .-
 - (a) Uncontrolled greenhouse temperature, (60).
 - (b) Greenhouse-thermostatically controlled at 70 F. (70).
- 3. Four species of plants,-
 - (a) Amelanchier alnifolia. (A. alnifolia).

 - (b) Cotoneaster acutifolia. (C. acutifolia). (c) Lonicera tatarica. (L. tatarica).
 - (d) Syringa villosa. (S. villosa).
- 4. Four acids,-
 - (a) Indole-acetic acid. (1).
 - (b) Indole-propionic acid. (2).
 - (c) Indole-butyric acid. (3).
 - (d) Naphthalene-acetic acid. (4).
- 5. Three concentrations,-
 - (a) 0 concentration, serves as a check. (0).
 - (b) 20 mg./100 cc. H20. (20).
 - (c) 40 mg./100 cc. H₂0. (40).
- 6. Basal cut at two places,-
 - (a) At the node. (A).
 - (b) One-quarter to one-half inch below the node, depending upon length of internode. (B).

Using Tippitts' tables of random numbers these variables were randomized in the order listed above. The replicates were first randomized in the frames, the two types of medium randomized within the replicates, then the two temperatures within the media, and so on, till lastly the two types of cuts occur within all the variables. After this, the rooting frames were blocked off in such a manner as to give 768 small areas corresponding to the number of randomized component variables. Each individual space was large enough to

^{*} Wherever symbols have been used in the tables, appendix or discussions they correspond to those shown in brackets after the above variables.

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accommodate 5 cuttings. Thus, the total number of cuttings required for the experiment was 3840 or 960 cuttings of each species. Strips of glass were inserted in the medium, to separate treatment from treatment.

A complete plan of the experiment was drawn on paper. The purpose was to facilitate planting and to do away with the necessity of labelling each small area.

Maturity of the Cuttings.

In the propagation of greenwood cuttings, the proper stage of maturity is a very important and essential factor. It is difficult to describe the proper condition or tell when this stage will be reached, since it depends largely upon environmental conditions. It will be necessary to take cuttings later than usual in seasons unfavorable for rapid new growth, or when abundant rainfall prolongs growth and delays maturity. Conditions favorable for early growth, with a lessening of moisture early in June, to encourage a partial ripening of the wood, bring the cutting material to the right stage by about June 20. Of course, the length of time over which the cuttings may be taken will depend upon the kind of season, since conditions which prolong growth will enable the material to be taken later than when the wood becomes mature early. The hardened, well-matured wood does not seem to respond so readily to root-stimulating substances as does newer growth.

Gathering the Cuttings.

The 1937 experiment was not designed to study the question of maturity. Only one lot of cuttings was taken, commencing July 19. The main reason for taking the cuttings at this late date was the slowness of growth in the spring of 1937. As far as possible, only wood of uniform maturity was selected. The cuttings were gathered

• grave to the state of the state With the second of the second ing the first term of the fir in the early part of the day, generally around 9 A.M., while still quite turgid and before any wilting had occurred. Only one cutting, the uppermost 5 - 6 inches of the new shoots, of the current season's growth was used. As soon as taken, the cuttings were placed in a bucket partially filled with water. This was a precaution against excessive wilting, because once a cutting has wilted severely it is difficult to bring it back to normal.

Preparation of the Cuttings.

After gathering, the material was taken to the potting shed where it was prepared. Here, with the aid of a sharp knife, many of the lower leaves were trimmed off, and the cutting shortened to the desired length, which was approximately 5 inches, depending somewhat upon the length of internode. The question of the number of leaves to trim off is one that is difficult to answer with any degree of accuracy. To some extent it will have to be learned by experience. Since the greenwood cutting has little or no storage of available food supply for root development, as much leaf area as possible should be retained, and yet the leaf area must not be so large as to cause excessive transpiration and wilting. The object should be to strike a balance between these two conditions. Half of the basal cuts were made by a transverse cut at the node, while the other half were cut transversely one-half inch below. The cuttings were then tied in very loose bundles and treated. Better and more uniform absorption of the chemical is secured when the bundles are not made too large or tied too tightly.

Treatment of Cuttings.

The four acids used, as root-promoting substances, were in crystalline form, and before using had to be made up into solution.

Most plants respond to very dilute concentrations only and, in

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addition, many plants possess a narrow range of concentration over which the phytohormone gives the desired stimulating effect.

This makes definite and accurate concentrations of utmost importance. In order to facilitate measuring the proper concentrations a stock solution of each acid was made up. A definite amount of acid,

200 mg., was weighed out, placed in a small beaker, and 1 c.c. of

95 per cent ethyl alcohol added to dissolve the crystals. A little

warm distilled water was added and thoroughly stirred, and then

made up to 100 c.c. of solution. Distilled water was used throughout the treatments. The stock solutions were kept in closely
stoppered bottles stored in a cool, dark place. The desired concentrations were obtained as required by diluting a portion of the

stock solution. Fresh solutions were made up for treating each
species.

Treating the Cuttings.

After the cuttings were prepared, they were placed in three series of beakers containing the three concentrations of phytohormones. The first series contained only distilled water and no acid, and will be termed 0 concentration. This concentration serves as a check, and was worked in with concentrations rather than treatments as this arrangement made it possible to fit the randomized variables into the frames more conveniently. The next series of beakers contained a concentration of 20 mg. per 100 c.c. of water and the last series contained 40 mg. per 100 c.c.. Each series required four beakers, to accommodate the four acids being used. Enough solution was added to cover \(\frac{3}{4} - 1\) inch of the lower portion of the cuttings. The beakers were then placed in a shaded part of the room and shielded from draughts.

The atmospheric conditions surrounding the soaking cuttings

is a matter of some consequences. High temperatures, and more particularly warm draughts, cause increased evaporation from the leaves with a resulting increased uptake of solution. On the other hand, a lower temperature, high humidity, with little or no air movement, greatly reduces evaporation, and consequently less solution is taken in by the cuttings. This variation in uptake of solution is, more often than is suspected, a cause of failure due to over- or under-dosage. As far as possible, this factor was eliminated by holding conditions nearly alike. Crowding in the beakers should be avoided, as this also reduces evaporation. Planting the Cuttings.

hours, before planting in the rooting frames. At the expiration of this time, they were removed from solution, washed in tap water, placed in a fresh beaker of water, and taken to the frames for planting. As was previously mentioned, a plan of the experiment was prepared. This plan was followed when inserting the cuttings in the medium in their allotted places. A hole was made with a dibble, the cutting inserted about two-thirds of its length, and then well firmed in. Care was exercised, at this point, to make

sure that the buried portion of the stem was firmly surrounded by

the medium to prevent drying out. After planting, the cuttings

The cuttings were allowed to stand in the solution for 24

Care of the Cuttings in the Frame.

were watered thoroughly.

Proper care of the cuttings in the rooting frame is a factor of profound importance. A little neglect at this stage will soon upset the results of the most carefully planned experiment. The necessity for proper care is obvious, when it is borne in mind that the softwood cutting has little stored food material to draw

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upon for root development, and that as little wilting as possible should be permitted until roots have developed and capable of drawing moisture from the medium.

The object should be to provide as much sunlight as possible without causing serious wilting. After the cuttings were planted and well watered in, the foliage was sprinkled with a very fine spray, and the sash closed down tightly for a time to secure a high humidity. The frames were shaded from too bright a light, for a day or so. After the cuttings had become readjusted to the change in environment, shading was withdrawn except during bright sunlight which tended to cause further wilting. Once rooting began, the amount of sunlight was increased.

The aim should be to keep the material in a turgid and fresh state. After the cuttings had been in the frame about one day the sash was raised slightly to allow for a change and circulation of air in the frames. Three or four times daily, the cuttings were syringed with a fine spray. Also, close observation was maintained for any signs of disease. Once or twice mold started on decaying leaves, and, when found, such leaves were immediately removed, the amount of sprinkling reduced to a minimum consistent with the health of the cutting, and more sunlight permitted.

From time to time the rooting medium was examined for moisture content and when necessary more water was added. The objective was to maintain the moisture content at 60 to 65 per cent. During hot, sunny days the frames and walks were sprinkled frequently, in order to keep up the humidity, and to maintain a fairly constant temperature.

Results.

Beginning after the third week, a few cuttings were lifted at weekly intervals to observe the progress of rooting. Usually in

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four to six weeks they were well rooted and ready to remove from the frames.

To indicate the rooting response of the cuttings to the phytohormone treatments, and to the other factors being tested, counts
were made of the number of cuttings producing roots. In addition,
the actual number of roots produced per cutting was counted. These
data were then used as the indication of the rooting response. The
average length of roots produced was estimated as closely as
possible, by a system of ranking, but this information has not been
included in the data, because of breakage to the longer roots in
removing them from the medium. The error was too great to justify
drawing any conclusions on this point.

Summary sheets for the data on the number of cuttings rooted and the number of roots produced are shown in Appendices I and II respectively. The appendices do not include Amelanchier almifolia, because throughout the entire experiment not a single cutting of this species produced any roots. In order to facilitate applying an analysis of variance to the results, it was deemed advisable to omit this species. An analysis of variance was applied to the data, in Appendices I and II, and is shown in Tables I and II.

Selection of an Error to Test the Significance of the Individual Factors and Their Interactions.

An examination of the columns for "Degrees of Freedom" and "Variance" in Tables I and II shows that the residual variance is very small, and, obviously, should not be used as an error to test the significance of the individual factors or their interactions.

Also, the selection of any one single error would seem to be inadequate to test all the factors and interactions. The error that should be used will depend largely upon what factors are to be

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	D.F.	Sums of Squares.	Variance.	<u>E</u>	5%	78
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TABLE I (cont'd.)	D.F.	Sums of Squares.	Variance	(Er.)	20	1%
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Temp. x treat. x cuts	wa		4 1	1 1	1 1	1 1
x treat.	1,2,1	85.5	1	6.85*	1.88	2.42
Spec. x treat. x cuts Spec. x concen. x cuts	0 4		50		1 1	1 1
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	70					
Ħ	9		0	1	9	8
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* Indicates the factors and interactions which exceed the 1 per cent point.

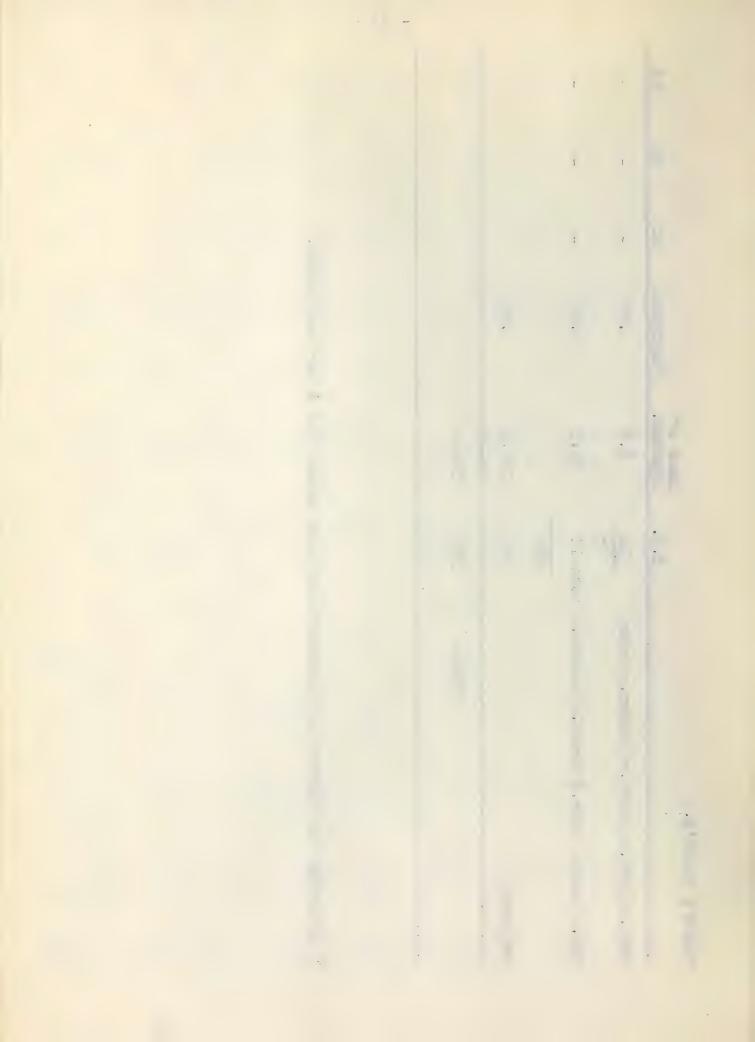


TABLE II. Analysis of Variance for Number of Roots Produced during the 1937 Experiment.

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perature cies atments centrations	_	3,122.	3,122.0	1	1	1
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atments centrations s	2	3,784.	1,892.3	9.48	0	-
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M	n	,855.	,951.7	5.31*	2.70	3.98
Med. x concen.	2	86.	43.3	•	*	
Med. x cuts	٦	116.	16.5	1	1	
Temp. x spec.	2	,708.	,854,4		3.10	∞
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×	01	7,756.	3,878.1	.04	-	
Temp. x cuts	<u>,-1</u>	381	381.9	1	1	1
Spec. x treat.	9	436.	239.4	8.15*	2.16	2.93
×	4	13,604.	8,401.0	.47	4	4
pec. x cuts	W.	2,755.	1,377.9	1	1	1
Treat. x concen.	9	,050.	6,008.4	11.56*	2.16	2.93
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Concen. x cuts	2	,167.	583.8	1	1	1
M	c)	,123.	,061.6	1	1	1
x temp. x	n	1,929.	643.1	1	1	1
H	2	,850	25.1	5.54*	3.20	5.10
×	-	781.	781.7		1	1
×	9	,358.	26.4	2.41	2.24	3.10
Med. x spec. x concen.	4	3,786.	46.5	1	1	1
H	W.	,094.	47.0	4.91	3.20	5.10
x treat. x	9	4,860.5	810.08	1	1	
. x treat. x	n	,428	76.3	1	•	1
x concen. x	a,	,598.	99.3	1	1	ŧ
Temp. x spec. x treat.	0	,540.	90.1	1	1	1

II (cont'd.)

TABLE

Indicates the factors and interactions which exceed the 1 per cent point.

tested, and what information is desired. The main purpose of determining the F values, shown in the analyses tables (Table I and II), was to obtain some indication of the more important variables and their interactions, and not the true F value. The method of selecting an error, which seemed the most adequate, was to group all higher interactions containing the variable or combination of variables, that was to be tested, and use the mean square of the group as the error. For instance, to test the significance of the results obtained for different species all interactions containing species were grouped, and the resulting variance used to determine the F value for species. Similarly, to determine the significance of the first order interactions all higher order interactions, which contain the two factors in the first order, are grouped and the mean square of the grouped total used as an error to determine the F value of the first order.

It was by this means that the F values, shown in Tables I and II, were secured.

Discussion of the Results.

In the discussion of the results the chief aim will be to point out the effects produced by the individual factors being studied, and how they have interacted to influence the rooting response. No attempt will be made to extract all of the information from the analyses and data, but only those conclusions will be drawn, that seem of most practical value. Each individual factor will be considered, followed by the important interactions involving that variable.

Media.

The differences exhibited (see Tables I and II) by the two types of media are insignificant. This would indicate that it

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makes little difference which form of medium is used insofar as the total number of cuttings rooted or the total number of roots produced is concerned. However, the effect of the two types of media is greater than the above statements would indicate. It will be observed that media interacts with treatments to give an F value greatly in excess of the 1 per cent point. The corresponding interaction for number of roots produced (Table II) is even more significant than for the number of cuttings rooted. Thus, it seems that the results for media vary with different treatments. Table III illustrates this point.

In treatments with acids 1 and 2 more cuttings rooted, and also more roots were produced in the sand than in the mixture. In treatment 3, the sand-and-peat medium was preferable, while no differences occurred between the two types of media in treatment 4. These results seem to suggest that where acids 1 and 2 are to be used then sand is the preferable medium, but that sand-and-peat may be expected to give better results when acid 3 is used. It is difficult to say, without further study, just why the different acid treatments should affect the results for the two media. Probably the above conclusions should be qualified by stating that such results could be expected again, only by using media of the same composition.

Basal Cut.

The question of the best place for the basal cut is one that has always been given much consideration in the older method of propagating the greenwood cutting. The analyses, for the 1937 data, show that the differences obtained are insignificant, whether it be for the number rooted or the number of roots produced. It would seem possible that such variables as species sensitivity

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TABLE III.

Treatments on the Number of Cuttings Rooted and Number of Roots Produced.

		534	193	727.
Totals		645 13,534	12,193	25,727.
Tot		645	609	1254
4	Roots	1,563	1,523	3,086 1254
	Rooted	118	118	236
Treatments 3	Roots	4,810	5,246	10,056
	Rooted	167	187	354
	Roots	2,013	1,811	3,824
2	Rooted	139	125	264
	Roots	5,148	3,613	8,761
1	Rooted	221	179	400
Medium		Ŋ	ا + ا	

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and treatments might influence or be influenced by the type of cut, in relation to the rooting response. However, under the conditions of this experiment, the analyses do not indicate any relationship with the other factors, so that within the limits of the species, treatments, concentrations, temperature, and media used, the place of the basal cut has made little difference.

Temperature.

The role of each of the remaining variables, temperature, species sensitivity, concentrations, and treatments, is more difficult to determine. Further examination of Tables I and II indicate that the above factors, as well as some of their interactions, are significant in themselves, but may or may not be significant in relation to one another. These variables will be discussed as they occur in the tables.

The results for temperature show a high F value, and at first glance one would be likely to conclude that temperature was highly significant in terms of the whole experiment. Closer examination of the first order interactions, reveals that treatments have interacted strongly with temperature, as is shown by the significant temp. x treat. interaction. A comparison of the mean square for temperature with the mean square for temp. x treat. gives an F value of 3.77, which is insignificant when based on 1 and 3 degrees of freedom. This indicates that any conclusions regarding temperature should not be based upon the results obtained for the different temperatures tested, but should be governed by the temp. x treat. interaction. That is, the best temperature cannot be stated, without first specifying the acid treatment to be used, because the statistical significance of the temp. x treat. interaction means that treatments have not responded in the same manner for all temperatures. This relationship is brought out in Table IV.

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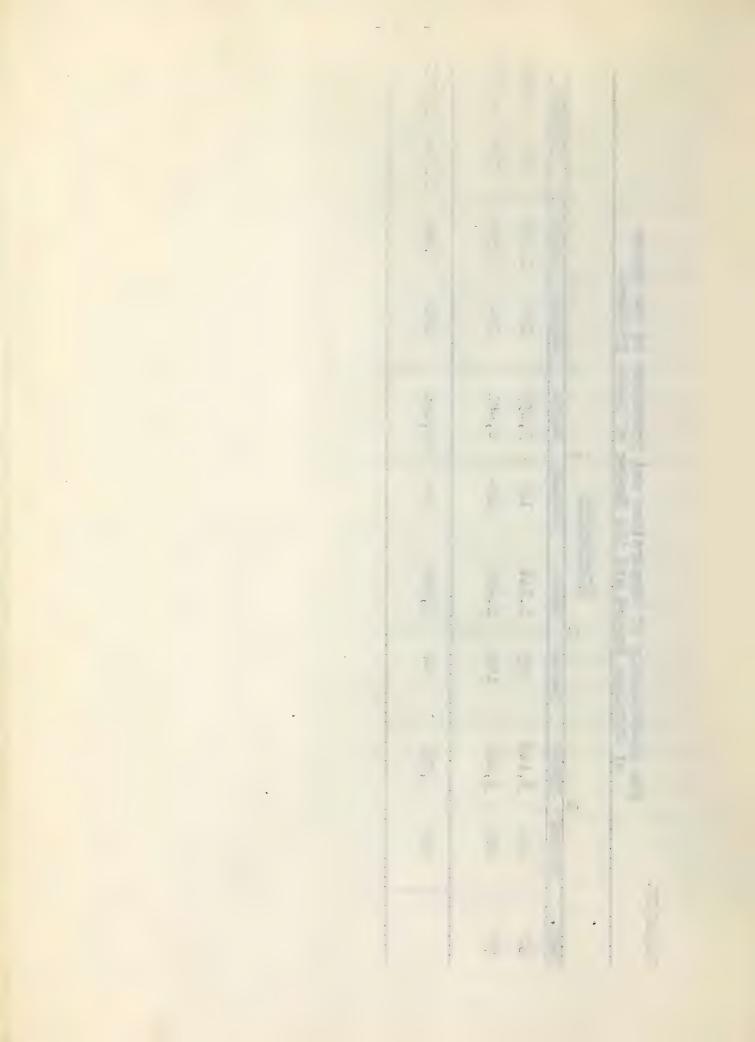
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TABLE IV.

The Relationship of Temperature and Treatment on the Number of Cuttings Rooted and the Number of Roots Produced.

	Totals	689,6	16,038	25,727	
	To	519	735	1,254	
4	Roots	1,520	1,566	3,086	
	Rooted	132	104	236	
Treatments 3	Roots	3,655	6,401	10,056	
	Rooted	140	214	354	
	Roots	1,366	2,458	3,824	
	Rooted	95	169	264	
-	Roots	3,148	5,613	8,761	
	Rooted	152	248	400	
	Temp.	009	⁰ 0L		



From the standpoint of cuttings rooted the greatest response was at 70° F. in treatments 1, 2 and 3, but the converse was true in treatment 4. The data in the above table shows why the total results, for the two temperatures, which favored 70° F., may not be used to indicate the better temperature of the two. Reference to Table II, for number of roots produced, shows that the same relationship holds between temperature and the temp. x treat. interaction as found in Table I. The number of roots produced in treatments 1, 2 and 3, at the different temperatures, seem to correspond proportionally to the number of cuttings rooted. In treatment 4 the number of roots produced has not been greatly influenced by the two temperatures of 60° and 70° F.

However, definite conclusions about the temperature and treatment relations should not be drawn from the data in Table IV without testing to see whether some of the other factors may not have influenced the temp. x treat. results. In fact, this is exactly what has happened, as shown by the significant temp. x treat. x concen. interaction (see Tables I and II). Statistically, the mean square for the temp. x treat. interaction is not significant over the mean square for the temp. x treat. x concen. interaction. For this reason, one cannot specify any best temperature for a given acid treatment without stating the concentration involved. Furthermore, conclusions should not be drawn from the second order interaction of temp. x treat. x concen., since its variance is not significantly greater than the variance of the third order interaction of temp. x spec. x treat. x concen.. Thus, to study the effect of temperature. and its relation to the other factors of the experiment, it will be necessary to refer to Table V. Study of the data in this table will show how the results, that helped to make up the first order inter-

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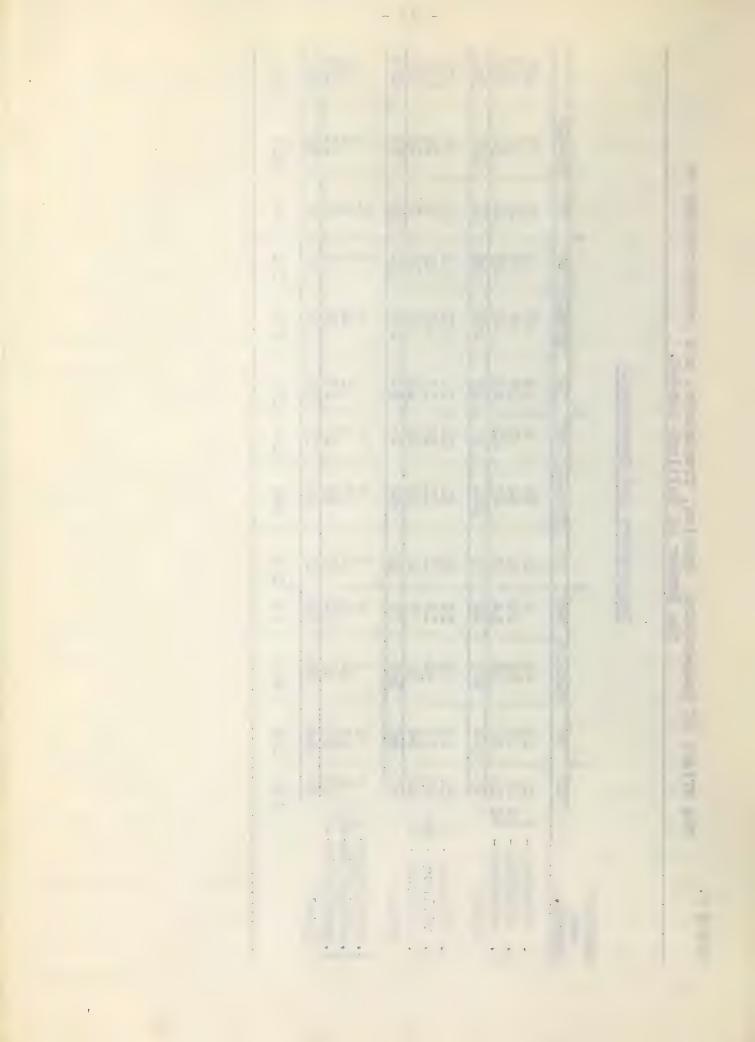
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The Effect of Temperature, Species, Treatments, and Concentrations on the Number of Cuttings Rooted.

Temperature and Treatments.

		148 217 202	9	171	486	119	201	1254
umand	Totals	37 41 50		40 29 20	89	1881	19	236
4	700	1.9 2.3 2.1		17	33	HW4	8	104
4	009	18	65	2021	56	01/0	11	132
	Totals	24 69 47	140	51 65 44	160	386	54	354
•	200	220		255	94	0 12 25	37	214
W	09	442	57	16 30 20	99	13	17	140
	Totals	53		21 20 20	92	17	40	264
(700	32	89	33	55	0000		169
N	09	200		108	37	1000	15	95
_	Totals	4 20	167	W R Z Z Z Z Z Z Z Z Z Z Z Z Z Z Z Z Z Z	145	0 0 4 4	ထ	400
	700	328	1,04		80	040	64	248
	09	127	63	220	65	1860	24	152
Species	Concen.	L. tatarica - 0 L. tatarica - 20 L. tatarica - 40	Total	급급급	Total	G.acutifolia-0 G.acutifolia-20 G.acutifolia-40	Total	



action of temp. x treat., were influenced by concentrations, and how the second order interaction of temp. x treat. x concen. has been influenced by different species. Table V points out clearly the interactions which occur among the four variables in question. Since this is the largest significant interaction involving temperature, all conclusions regarding temperature, and its relation to the other factors should be based upon the data in the table showing the interaction.

It will be noticed that a temperature of 70° F. has given better results in nearly every combination of species and concentrations, within treatments 1, 2 and 3. However, the advantage of 70° F. over 60° F. is not proportionally the same in all combinations of species, treatments and concentration. With the exception of S. villosa at 0 concentration, the 70° temperature has been the most advantageous for all the remaining combinations of species, and concentrations, within the first three acids. This would seem quite conclusive proof that, for indole-acetic, indole-propionic, and indole-butyric acids. 70° F. is the best temperature. In acid treatment 4 the above situation is not the case. Considerable interaction appears to be present within naphthalene-acetic acid (treatment 4). For all three concentrations the number of cuttings rooted, in S. villosa, has been definitely greatest at 60° F. The same situation appears to have occurred in C. acutifolia, but with L. tatarica the effect of temperature is less evident. In the latter species the results would seem to be strongly influenced by different concentrations. However, greatest rooting has been effected at a concentration of 40 mg. / 100 c.c. H20, at 60° F. Before these conclusions concerning treatment 4, and its interaction with temperature could be drawn with absolute confidence, it would

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be necessary to verify them by further experimentation.

Attention is drawn to the fact that the conclusions from the data for the temp. x spec. x treat. x concen. interaction (Table V), as regards the better temperature at the various treatments, do not differ greatly from those suggested when discussing Table IV. This close similarity is probably coincidental, and would not always be expected to occur between first and third order interactions. One important difference does occur, in that, the best temperature in relation to each treatment is stated in terms of each species and concentration, in the third order interaction, but this cannot be done from the first order. Statistically, the conclusions pertaining to temperature and treatments are only valid when species and concentrations are also considered.

The Effect of Temperature and Its Interaction on the Number of Roots Produced.

The relationships of temperature to the other factors, from the standpoint of number of roots produced, are similar to those for the number of cuttings rooted. Reference to Tables I and II shows that practically the same interactions are significant, and to approximately the same degree. Following the same method of testing one significant interaction against another, as used in discussing the temperature relations in Table I, it may be shown that the effects of different temperatures cannot be properly stated, without taking into account the combined effects of temperature, species, treatments and concentrations. Thus, any conclusions regarding the temperature relations must be taken from the statistically significant temp. x spec. x treat. x concen. interaction (see Table VI).

From the standpoint of the number of roots produced, temperature seems to be more variable in its relations to species,

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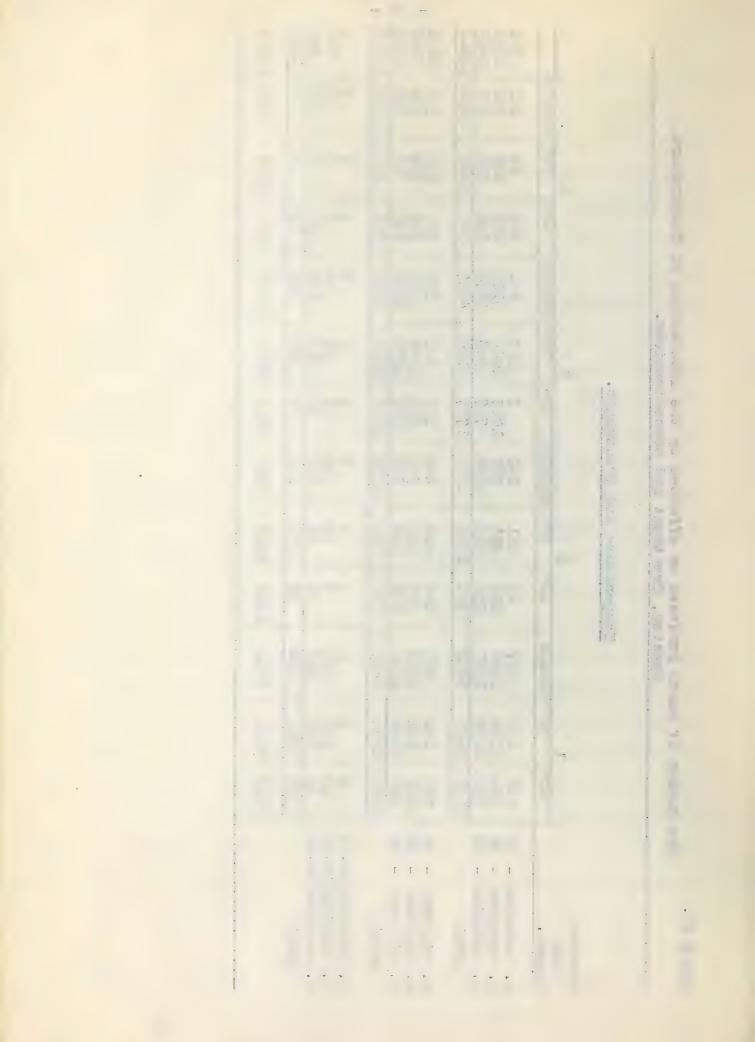
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TABLE VI.

The Number of Roots Produced as Affected by the Interaction of Temperature, Species, Treatment and Concentrations.

Temperature and Treatments.

Species		1			2			W			4		
concen.	209	700	Totals	209	00%	Totals	009	200	Totals	600	700	Totals	
· tatarica -	4		5	41	0	4	2	10	4	-	0.0		1 4
L. tatarica - 20	5	1194	1748	0	0	787	9	14	10	10	10	1 4) W
· tatarica -	392	M	9	254		00	707	963	1670	348	525	873	42
Total	∞	2613	3802	0	1522	2113	1777	3140	4917	741	00	30	12662
. villosa -	M	9	0	4	S	9	5	4	0		1	4	5
	1006	962	1968		228	557	811	14	IN	-0		t C	1 0 1 ∞
· villosa -	5	5	디	5			M	-	M	124	15	181	4609
Total	1901	2574	4475	732	801	1533	1824	2961	4785	9	463	24	12
. acutifolia-	0	0	0	0	0	C	C	C		C	0	C	
C. acutifolia-20	13	218	M	6	55	64		63		ο α) rc	7 12	α
· acutifolia-4	45	0	253	34	, , , ,	114	43	237	280	10	15	15	664
Total	58	426	484	43	135	178	54	0	15		14	32	
	3148	5613	8761	1366	2458	2824		5401	73001	C	L	700	0
)	-	+ 10			000	1250	1200	2000	1.21.62
								-					



treatments and concentrations (Table VI), than it was for the number of cuttings rooted. However, from the data in Table VI it will be observed that 70° F. has been more favorable in the majority of combinations of species, treatments and concentrations. In L. tatarica, at concentrations of 20 and 40 mg., and in all treatments. 70° F. gave consistently better results than did 60° F. relationship was not true in all the checks. C. acutifolia behaved in a similar manner to the former species, except that in treatment 4, 60° F. may be the more preferable temperature. With S. villosa the best temperature cannot be stated as definitely as with the other two. Before recommending the optimum temperature in the latter species it is necessary to know the treatment and concentration that are to be used. For instance, with indole-butyric acid the greatest response occurred at a concentration of 20 mg. and 70° F., whereas with naphthalene-acetic acid more roots were produced at 20 mg. and 60° F.

Further study of Table I shows that the results for temperature are definitely significant in relation to medium and cuts, and the interactions containing these two variables. From this it seems safe to conclude that regardless of the medium used, or the place of the basal cut, the 70° F. temperature may be expected to produce the greatest rooting response.

Species Results.

The results obtained for the three species tested are far beyond the possibility of these differences having occurred by random sampling. The species results are highly significant as is indicated by the large F value (Table I). After comparing the mean square for species with the mean squares for the significant interactions containing species, it is found that statistically the

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species results are significantly greater than all the interactions, with the exception of spec. x concen., to exceed the 5 per cent point. Thus, regardless of the medium, cuts, temperature, and the kind of acid used, the differences actually obtained for the different species are a true indication of the order of their ability to root. From the standpoint of number of roots produced (Table II) the variances for the species results are significantly greater than the variances for the interactions containing species, excepting the temp. x spec. and spec. x concen. interactions. Since the spec. x concen. interaction is common to the number rooted and the roots produced it will be discussed first.

Species and Concentration Relationship Affecting the Rooting Response.

The high F values for the spec. x concen. interactions (Tables I and II) indicate a very strong interaction between these two factors. It is to be expected that the different species respond differently to definite concentrations. The results of this interaction are shown in Table VII.

In <u>C. acutifolia</u> a progressive increase in rooting response has occurred with an increase in concentration from 0 to 40 mg..

Maximum rooting in <u>L. tatarica</u> was at the concentration of 20 mg..

S. villosa shows a gradual decrease in rooting from the 0 to 40 mg. concentration. At first glance, this might seem to indicate that if the acids used are really effective in stimulating this species, then the best concentration of acid would most likely be between 0 and 20 mg.. However, from Table VII it is impossible to conclude which concentration is best for the various species since the different acid treatments have influenced the results shown in Table VII. This condition is indicated by the significant spec. x treat. x concen. interaction, and also by the fact that the mean square

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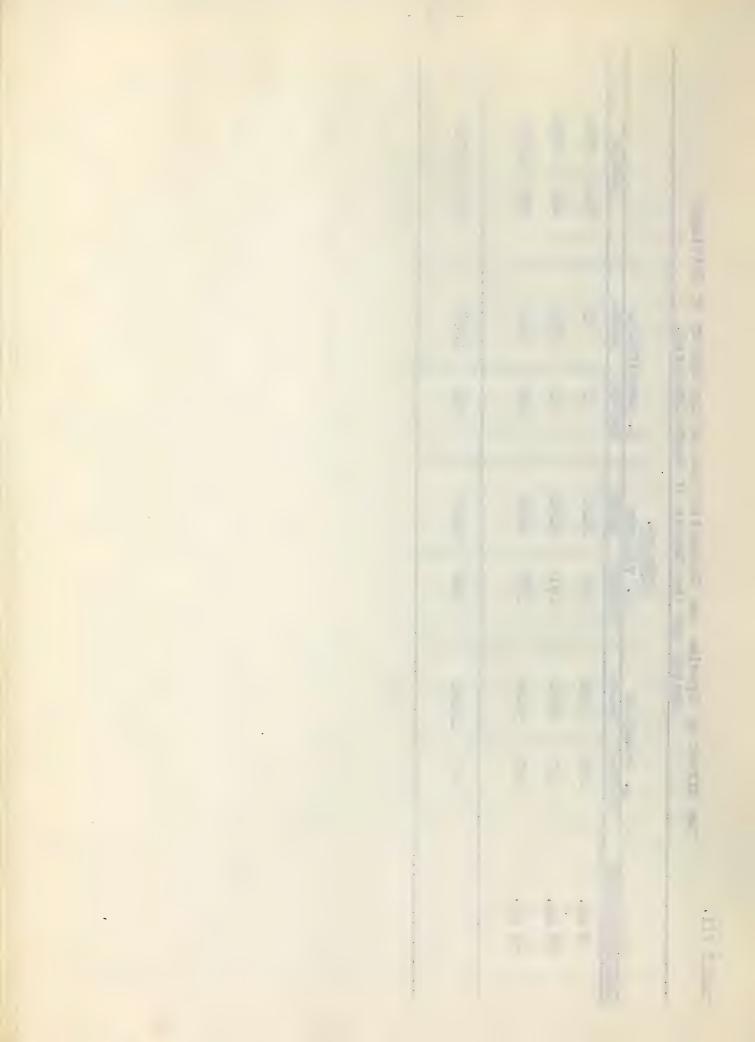
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TABLE VII.

The Effect of Species and Concentration on the Number of Cuttings Rooted and the Number of Roots Produced.

		0	0	7		
	Totals	2980	12050	10697	1254 25727	
	To	326	469	459	1254	
9	Roots	2	382	664	1048	
5	Rooted Roots	1	81	119	201	
Species.	Roots	2126	5282	4609	12017	
Spec	Rooted	177	171	158	486	
4	Roots	852	9829	5424	12662	
t at a t a t a t a t a t a t a t a t a	Rooted	148	217	202	567	
	Concentrations	0 mg.	20 mg.	40 mg.		



for the spec. x concen. interaction is significant in relation to the mean square for spec. x treat. x concen.. This makes it: apparent that before the best concentration can be stated for any species it is necessary to know which one of the four acids is to be used. Information on the spec. x treat. x concen. interaction may be secured by grouping the appropriate data from Appendices I and II. Even though this information were obtained any conclusions drawn therefrom would not be justified, since further examination of the analyses shows the spec. x treat. x concen. interaction to be insignificant in terms of the higher interaction of temp. x spec. x treat. x concen.. The significance of this third order interaction means that the species results as obtained, were influenced by temperature, by the acid treatments, by the concentrations, and by the conditions which have made one or other of the temperatures favorable or unfavorable at different treatments, and also the conditions which have made combinations of temperature and treatment advantageous or disadvantageous at different concentrations. Thus, it becomes apparent, from the foregoing discussion, that the best concentration for each of the three species must be stated in relation to a definite treatment and temperature. To secure this information regarding species it will be necessary to refer back to Tables V and VI.

A few important conclusions regarding the combinations of temperature, treatment and concentrations that seem best for each species will be enumerated. It will be noted from Table V that the greatest amount of rooting in <u>L. tatarica</u> is associated with treatment 1, using a concentration of 40 mg. at 70° F. However, this combination of factors is not the most favorable for the number of roots produced (Table VI). In the latter, treatment 3

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at a concentration of 20 mg. and 70° F. was definitely superior to all other combinations. The advantage of only one more cutting rooted, under the first set of conditions, probably means very little, so that the best combination of variables to recommend for L. tatarica would be the second set, i.e. treatment 3, concentration of 20 mg., and 70° F. The maximum rooting response in S. villosa was approximately equal at two different combinations of factors. Equally good results may be expected with treatment 1, at a concentration of 40 mg., and 70° F., and with treatment 3, at 20 mg., and a temperature of 70° F. In C. acutifolia the greatest amount of rooting resulted from treatment 1 at 20 mg., and 70° F. A slightly greater number of roots were produced with treatment 3, at 40 mg., and 70° F. The first combination of variables is probably the more preferable, since it has given the maximum rooting response.

The percentage rooted as well as the number of roots produced is greatest in L. tatarica, followed in order by S. villosa, and C. acutifolia. Rooting of C. acutifolia cuttings was markedly stimulated by phytohormone treatment.

Temperature x Species Interaction.

The significant temp. x spec. interaction in Table II, which was referred to earlier in the discussion of species results, indicates a high degree of interaction between temperature and the different species. It will be remembered that the differences for species were insignificant in relation to the temp. x spec. reaction, which meant that the order, in which the different species responded most readily, would not necessarily be the same in which they would occur at different temperatures. Although significant, the temp. x spec. interaction is not sufficiently greater than the temp. x spec. x concen. interaction to exceed

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the 5 per cent point. Thus, it appears that concentrations have strongly influenced the results of the temp. x spec. interaction, and it cannot be concluded safely, as to which is the better temperature for the different species, without considering the concentration. In addition, the different treatments have affected the results for the third order interaction, temp. x spec. x concen.. Statistically, the variance for temp. x spec. x treat. x concen. is sufficiently greater than temp. x spec. x concen. to make it necessary to draw all conclusions, regarding the temperature and species effects, on the number of roots produced, from the third order interaction, temp. x spec. x treat. x concen., as shown in Table VI. The conclusions have been drawn already, in connection with this table, which involve the temp. x spec. relationship now being discussed. It becomes apparent from Table VI that any best temperature cannot be stated, that will apply to all species. Treatment and Concentration Relationships.

Some of the effects produced by the different treatments and concentrations have been suggested while discussing the relationship of the other variables. The individual factors of treatment and concentration are highly significant in themselves. The differences obtained for each variable were sufficiently great to ensure that if the entire experiment were repeated the same differences would be expected to occur in at least 99 times in 100 trials. In spite of this, it should not be concluded that the totals, as obtained for the various treatments and concentrations, provide a true indication of what treatment or concentration are the best, for every condition within the experiment. This is supported by the fact that the mean squares for treatments and for concentrations are insignificant over the mean squares of the first

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order interactions of temp. x treat., spec. x concen., and treat. x concen. The necessity of considering treatments and concentrations in relation to temperature, species, and their own interaction, becomes apparent from the above remarks. In the previous discussion of temperature, and species, it has been shown that to discuss the temp. x treat., and spec. x concen., relations it was necessary to consider the effects of treatments and concentrations. Similarly, it may be shown statistically, that the treat. x concen. interaction should only be discussed in terms of the different species, and temperatures. This simply means that, before recommending a treatment or a concentration, it would be essential to know what species are to be used and at what temperature. any conclusions involving the above combination of factors will have to be based upon the significant temp. x spec. x treat. x concen. interaction (see Tables V and VI). A few of the general practical recommendations have already been drawn from these tables, during the discussion of species results, and it will be unnecessary to enlarge upon, or repeat, these conclusions at this point.

Throughout the discussion of results the aim has been to indicate the effect of each variable and combination of variables on the rooting response of cuttings. It is evident that the number of cuttings rooted and number of roots produced is the result of the interaction of all the variables and is not due to one or two acting independently. The combination of factors that has encouraged maximum root production is shown in Table VIII.

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TABLE VIII.

The Combination of Factors which Produced the Maximum Rooting Response in the 1937 Experiment.

	Med	Medium	Temp.	Temperature	Treatment	ment	Conce	Concentration	Co	Cuts
Species	Number Rooted	Number of Roots	Number Rooted	Number of Roots	Number Rooted	Number of Roots	Number Rooted	Number of Roots	Number	Number of Roots
Cotoneaster	S OF C	S or	700L	700F	Indole- acetic	Indole- acetic	20 mg./ 100 cc.	20 mg./ 100 cc.	A or B	A or B
					3	3400				
Lonicera	S OF	S OF	70°F	T00L	Indole- butyric	Indole- butyric	20 mg./ 100 cc.	20 mg./ 100 cc.	A or B	A or B
					acıd	acid				- ;
Syringa villosa	S or	S or	700F	TooL	Indole-	Indole-	20 mg./ 20 mg./	20 mg./	A or B	A or B
	!				acid	acid	999	*000 CCC		•



1938 EXPERIMENT.

The 1938 experiment was conducted along similar lines to the 1937 one, and had as its purpose much the same objectives; namely, the study of type of medium, optimum temperature, place of the basal cut, ease with which different species root, the proper concentrations of phytohormones to encourage rooting, and the various interactions between these variables. A number of points, which arose out of the 1937 experiment, led to a few changes for the 1938 plan. Two additional factors, (a) the most suitable date to gather the cuttings, and (b) the question of rooting response under outdoor conditions, were included in the 1938 experiment. These will be discussed more fully under "Materials" and "Methods."

Materials.

Plant Materials.

possible, the number was increased from four to six. This was all that could be worked into the experiment conveniently. The six species tested were Cornus stolonifera, Corylus cornuta,

Rosa sp., Spiraea flexuosa, Syringa vulgaris, and Viburnum trilobum.

Because of a lack of sufficiently uniform cutting material, several otherwise desirable species had to be left out. As far as possible, cutting material was obtained from plants on the University grounds.

Spiraea flexuosa was supplied by Taylor's Nursery, South Edmonton.

The material of Cornus stolonifera, Rosa sp., and Corylus cornuta was obtained from natural habitats located a short distance west of the University along the banks of the North Saskatchewan river.

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Media.

The types of media tested in the 1938 experiment were similar to those employed in 1937. The grade of materials, preparation of the mixture, steam sterilization, and the method of handling the media were the same as in the previous investigation. Prior to planting any cuttings, pH determinations were made which showed sand to be slightly alkaline (pH 7.6), and the sand-and-peat mixture to be nearly neutral (pH 7.3).

Hormone Materials.

Most commercial nurserymen, as well as amateurs, interested in plant propagation with the aid of root stimulating substances, do not have the facilities to weigh out the crystalline form of these substances in the very minute quantities that are required. In order to secure satisfactory rooting it is absolutely essential that the proper concentrations be used, and this requires accurate balances. For these reasons, it was decided to use a commercial product in the 1938 experiment, in order that any information obtained might be of more practical value to propagators. Only one phytohormone was used, and that was Hormodin A, a product of the Merck Chemical Co., Montreal, Canada. In this preparation the organic acid, indole-butyric, is already in liquid form, and this facilitates measuring the proper concentrations.

Frames.

The same rooting frames as used in the previous experiment were again employed, but, in addition, two similar frames were placed out-of-doors in order to test the rooting response under outside conditions.

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Methods.

The experimental method followed was essentially the same as used the previous year. However, one or two changes were made in order to secure information on the response of cuttings taken at different dates, and the practicability of using outdoor conditions. In all, the following factors were given consideration: type of medium, temperature relations, species, concentration of phytohormone, place of basal cut, and appropriate time of year to take cuttings.

The 1937 work was conducted under greenhouse conditions, and as such facilities are not always available to those interested in this phase of plant propagation, it seemed advisable to study the response under outside conditions. Therefore, the previous year's plan was expanded in 1938 to include two frames, of the same general type as used inside, and these were arranged in a small enclosure outdoors. These frames were protected on the north by a caragana hedge, but exposed on the remaining three sides. In order to have the light relations as nearly identical as possible, the long axis of the frames was placed to correspond with those inside, namely, running north and south. This plan insures, insofar as possible, uniform sunlight to all parts of the frame. This is not the case when the long axis extends east and west. The frames were equipped with close-fitting sash, and shade was provided when necessary to reduce the amount of sunlight.

Attention is drawn to the fact that, what is referred to as temperature, in the analyses and discussion, is not temperature in the most strict sense. Rather, it refers to combinations of temperature and atmospheric conditions. Three sets of

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conditions were studied in this experiment. These were: (1) in the greenhouse with no definite temperature control, (2) in the greenhouse with the temperature thermostatically controlled at 70° F., and (3) under prevailing outdoor conditions. In the first and last conditions the temperature fluctuated somewhat with atmospheric changes; though by proper shading, and sprinkling with water, sudden or wide fluctuations were reduced to a minimum. It is realized that these are really three sets of conditions rather than three temperatures, but for want of a better term, "temperature" will be used. Each condition could have been considered as an individual experiment and analyzed as such; but, since it was desirable to make comparisons between inside and outside results, the three conditions have been included in one experiment.

Hormone Treatment.

As indicated under "Materials," only one phytohormone was used. The threefold purpose in doing this was; first, to obtain information of more practical value to nurserymen by using a more convenient form of hormone, second, to provide for the consideration of more species, and third, the best time at which to take the cuttings. Although it was realized that repeating the 1937 experiment, using the four crystalline acids, would greatly enhance the value of the results and conclusions, it was decided to forego this in order to study the three above mentioned factors, as they seemed of paramount importance.

The phytohormone solution was prepared in accordance with the directions accompanying the Hormodin A product. These directions facilitate accurate measurement of the desired concentration.

Three concentrations were included in the 1938 tests and consisted

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of 0, 20 and 40 B.T.I. units. The 0 concentration contained no acid and really served as a check.

Time of Season to Gather Cuttings.

To secure further information on the best time of year to gather the material, provision was made to include this variable. The cuttings were taken at three different dates as follows; (a) the first lot commencing June 20th, (b) the second lot about July 15th, and (c) the third lot starting August 8th. Throughout the tables and discussion these times of the season will be referred to as "dates."

Position of Basal Cut.

As in the previous work, the relative advantages of the two types of basal cuts were investigated. The two types of cuts,

(a) at the node, (b) one-half inch below the node, were again adopted.

Experimental Plan.

The complete experiment was designed for statistical analysis and randomized in a manner similar to that of the 1937 experiment.

Also, the experiment was done in duplicate. Details of the design were as follows:

- 1. Two types of media,-
 - (a) Sand. (S)*
 - (b) Sand + Peat. (S+P)
- 2. Three temperatures,-
 - (a) Greenhouse (Uncontrolled) Approx. 60° F. (60°)
 - (b) Greenhouse 70° F. (70°)

⁽c) Outdoor conditions. (Out.)

^{*} Throughout the appendix, tables and discussion symbols may often be used to represent the variables being studied. These are shown in brackets following the variables.

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- 3. Six species,-
 - (a) Cornus stolonifera. (C. stolonifera)

(b) Corylus cornuta. (C. cornuta) (c) Rosa sp. (R. sp.)

- (d) Spiraea flexuosa.(S. flexuosa)(e) Syringa vulgaris.(f) Viburnum trilobum.(V. trilobum) (d) Spiraea flexuosa.
- 4. Treatment .-

Hormodin A.

- 5. Three concentrations .-
 - (a) 0 concentration (check). (0)
 - (b) 20 B.T.I. units. (20)
 - (c) 40 B.T.I. units. (40)
- 6. Basal cut at two places,-
 - (a) At node. (A)
 - (b) One-quarter to one-half inch below node. (B)
- 7. Three dates in the year.
 - (a) June 20. (1)
 - (b) July 15.
 - (c) August 8. (3).

The same form of randomization, as previously outlined, was followed and the frames were blocked-off as before. The number of cuttings that could conveniently be planted in each individual block was four. This number was not as large as might have been desired, but when it is considered that the complete experiment required a total of 5184 cuttings, the results should be a pretty fair indication of the reaction of the different species to the different factors being studied.

Care and Handling of the Cuttings.

The manner of gathering, perparing, treating, planting, and caring for the cuttings was essentially the same as that followed in the 1937 work.

Rosa sp. is probably R. acicularis.

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Results.

In four to five weeks the cuttings were usually well rooted, and ready to remove from the frames. Sometimes the production of new growth was an indication that roots had developed, but this sign could not always be relied upon. It was more satisfactory to remove a few of the cuttings occasionally to see whether roots had been produced. As the cuttings were taken from the frames, the number rooted and the number of roots per cutting were recorded, and these data were used to indicate the final response to the factors being studied. As in the 1937 work, the average length of roots produced was estimated, but has not been used because the end of many of the longer roots broke off when removing the cutting from the medium. The error involved, seemed too great to justify recording and analyzing the data. The remaining data are shown in Appendices III and IV. These data were subjected to an analyses of variance test and the results of this analyses for the number of cuttings rooted and number of roots produced are shown in Tables IX and X, respectively.

The method of deriving the F values shown in the analyses was essentially the same as for the analysis of the 1937 data, excepting that the residual was included when grouping the interaction to use as error. This method gave a truer indication of the significance of the interactions.

Discussion of Results.

Throughout the discussion of results the chief aim will be to extract from the data the information and conclusions that seem of most practical value. Each individual factor will be dealt with first and followed by a discussion of its relationship to,

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TABLE IX.
Analysis of Variance for Number of Cuttings Rooted during the 1938 Experiment.

a). F.	Sums of Squares.	Variance.	janj	59	1
, to the total of	-		C		1	
Medium		4	4	4	00	6.66
Temperature		38.9	9.4	1.29	0	9
Species	100	,5	39.1	95.27*	2.22	0
Concentration	, cu	36.3	8.1	6.44	0.	9.
Cuts	 1	0.0	0.0	8		
Dates	N	9.1	.5	152.89*	3.00	4.63
Med. x temp.	2	14.1	0.	0.73	.0	9.
Med. x spec.	2	3.5	9		1	1
Med. x concen.	્ય	F-,	3	8	1	1
Med. x cuts	1	9.	9,	1	1	1
Med. x dates	N	1.2	9.0	1		1
Temp. x spec.	10	S	00	18.66*	1.86	2.37
M	4	2	5	3	ŧ	1
Temp. x outs	N	7.0	0.5	1		1
Temp. x dates	4	42.8	0.7	14.33*	2.38	3.34
H	10	7.0	-	5.18	œ	•
M	2	5.2	1.0	1		1
x da	10	1	٦,	35.42*	1.86	2.37
n. x	2	0.3	0.1	*	3	1
en.	4	3	5.2	14.67	2.38	3.34
Ħ	N	4.3	-	5	0	9
×.	10	• 4	0	.16	00	S
· x temb. x	4	6	3	1	1	1
. x temp. x	N	٠ 7	3	t	1	1
H	4	∞	• 4	1	•	1
x spec x	10	0	S	1	1	1
x spec. x	5	ر د 2	4	1	1	1
x spec. x da	07	4.	2	3.50*	1.86	2.57
x concen. x	· v	<i>y</i> (40	t'	t	2
Med. X concen. X dates	40	2,6 2,7 2,7 2,7	01.0 70 L	9 1	1 -1	8 1
an v cons v •	U	,	V	•	•	•

TABLE IX (cont'd.)

1%	1.94	
50	1.60	
<u>E</u>	2.26	
Variance.	00001100000000000000000000000000000000	
Sums of Squares.	1 4 1	3250.00
D.F.	01.00 101 010 101 040 0040 0444	1295
	Temp. x spec. x cuts Temp. x spec. x dates Temp. x spec. x dates Temp. x concen. x dates Temp. x concen. x dates Temp. x concen. x dates Spec. x concen. x dates Med. x temp. x spec. x concen. Med. x temp. x spec. x dates Med. x temp. x concen. x dates Med. x temp. x concen. x dates Med. x temp. x concen. x dates Med. x spec. x concen. x dates Temp. x spec. x concen. x dates Med. x temp. x spec. x concen. x dates Med. x temp. x spec. x concen. x dates Med. x temp. x spec. x concen. x cuts x dates	Total

* Indicates the interactions which exceed the 1 per cent point.

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Analysis of Variance for Number of Roots Produced during the 1938 Experiment. TABLE X.

	D.F.	Sums of Squares.	Variance.	도네 *	5%	1%
	-	111	ויווי.	3		
Medium	 	1.525	1,525.4	6.73	∞	9.
Temperature	03	45,914.	22,957.4	13	3.00	4.63
Species	r	66,380.	3,276.0	φ α	2	0
Concentrations	ณ	701,069.	50,534.5	25.13	•	9.
Cuts	 1	2,000.	2,000.1	1	1	1
Dates	2	,890.	,945.4	87.91*	3.00	4.63
Med. x temp.	ત્ય	4,911.	2,455.5	1	1	
Med. x spec.	2	,903.	,580.7	500	2.25	3.04
×	ત	4,594.	,297.1	1	1	1
Med. x cuts	,	51.	51.3	1	1	
Med. x dates	2	10,077.	5,038.9	0	3.00	4.63
Temp. x spec.	10	2,353.	,235.4	6	0	5
×	4	,269.	,567.3	6	n	3
Temp. x cuts	N	2,506.	,253.4			1
×	4	19,984.	4,996.2	1.00	S	3
H	10	616.	761.7	45.66*	1.86	2.37
H	2	17,123.	,424.8	2.30	2	0.
M	10	8,540.	,854.0	0	00	w
n.	2	634.	317.4	1	1	1
Concen. x dates	4	,877.	,469.4	18.47*	2.38	3.34
Cuts x dates	ત્ય	2,851.	,425.8	1	1	1
Med. x temp. x spec.	10	,830.	683.0	2.43*	1.86	2.37
· x temp. x	4	5,406.	,351.5	1	1	1
Med. x temp. x cuts	a	384.	192.0	1	1	1
Med. x temp. x dates	4	8,626.	156.5	1	1	1
Med. x spec. x concen.	10	,642.	,264.2	2.18	1.86	2.37
Med. x spec. x cuts	יכ	2,588	517.6	ŧ	1	1
× ×	10	2,879.	87.9	1	1	1
Med. x concen. x cuts	21	927.	463.7	1	1	1
Med. x concen. x dates	4	,143.	85°	1	1	1
Med. x cuts x dates	N	,633.	816.6		1	1
x spec. x	20	73,403.4	3,670.17	2.39*	1.60	1.94
x spec. x	10	5,340.	554.0		1	ı
Temp. x spec. x dates	20	,071.	53.5	1.83	1.60	1.94

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7	1 1 1	1.94	1 1 1	1 1 1	2.37	1 1 1	1.60	1 1 1 1	1 1 1 1	•	
5%	1 1 1	1.60	1 1 1	1 1 1	1.86	1 1 1	1.30	1 1 1 1	1 1 1 1	1	
<u>후</u>	1 1 1	11.70*	1 1 1	1 1 1	2.05*	1 1 1	1.51	1 1 1 1	11	1	
Variance.	90.3	10.9	,469.6 059.2 733.8	260 200 200 200 200 200 200	009 000 400.0	127.6	11,5875,70	,146.4 932.0 969.2	1400 1400 1400	91.2	
Sums of Squares.	961	335	7,878	2527	,561 ,608 ,811	4,147.	91,012.0	676 679 670 670 670	44 % % % % % % % % % % % % % % % % % %	650	8,156,995.9
D.F.	4 0 4	1001			200		240	0000		es 4 64	1295
	Temp. x concen. x cuts Temp. x concen. x dates Temp. x cuts x dates	x concen. x x cotts x dat	sen. x cuts x d x temp. x spe x temp. x spe	x temp. x spec. x temp. x concen.	Med. x temp. x cuts x dates Med. x spec. x concen. x cuts Med. x spec. x concen. x dates	N N N	x spec. x concen. x . x spec. x cuts x dat	x concen. x cuts x dates x temp. x spec. x concen. x c x temp. x spec. x concen. x d	x temp. x concen. x cuts x dates x spec. x concen. x cuts x dates x spec. x concen. x cuts x dates	emp.x spec.x concen.x cuts x	Total

* Indicates the interactions which exceed the 1 per cent point.

and interaction with, the remaining factors.

Media.

The results for the two types of media, in relation to the total variance for all interactions involving media, are signicicant to near the 1 per cent point (Table VIII). Yet, it is unsafe to draw any conclusions from the differences obtained, for it appears that the response to medium has been greatly influenced by temperature, as shown by the highly significant first order interaction, med. x temp.. This means that the results for media vary at different temperatures, so that sand, even though it gave the better results over the whole experiment, may not be the best for all temperatures. Furthermore, the relationship of medium and temperature has been partly brought about by the response of different species, as indicated by the highly significant second order interaction, med. x temp. x spec.. Statistically, the mean square for med. x temp. is not sufficiently greater than the mean square for med. x temp. x spec. to discuss the interaction of temperature and medium without first considering in what way species may have affected the results. The significant med. x temp. x spec. interaction means that media react differently at various temperatures for various species.

None of the higher order interactions, containing the combination, med. x temp. x spec., are sufficiently greater than the residual variance to be significant. This indicates that the remaining variables, concentrations, cuts, and dates, have not greatly influenced the med. x temp. x spec. interaction. The results for medium as affected by temperature and species will be more readily observed by reference to Table XI.

From the standpoint of the number of roots produced, the

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results obtained for media are significant (see Table X). However, the relationship of medium to the other factors differs somewhat from the situation for the number of cuttings rooted. In Table X the med. x temp. interaction seems of little importance, yet, medium did interact significantly, with species and with dates. The mean square for media is not significant over the mean squares for the med. x temp., and med. x temp. x spec. interactions. Since the results for media are in favor of sand, it would appear that for root production sand is the better medium at the temperatures tested. Even though sand has given consistently better results at all temperatures, the relationship between the two media at the three temperatures is not proportionally the same for all species. This is suggested by the significant med. x temp. x spec. interaction.

Further conclusions regarding medium and temperature relations, both as regards number of cuttings rooted and number of roots produced, must be based upon the data in Table XI.

For <u>S. vulgaris</u>, <u>V. trilobum</u>, and <u>C. stolonifera</u> sand at 70° F. has proved to be the better combination of medium and temperature. In <u>S. flexuosa</u> sand under outside conditions has been the most favorable, but in the remaining species the most advantageous medium and temperature combination is less obvious. If the objective is a balance between the greatest number of cuttings rooted and correlated with large number of roots produced, then sand-and-peat at 70° F. is probably preferable. Insofar as <u>R. sp.</u> is concerned there is little choice between sand, or the sand-and-peat mixture under outside conditions.

Relationship of Medium to the Other Factors.

The concentrations of Hormodin A, and the two basal cuts employed have had little effect on the results for medium. As far

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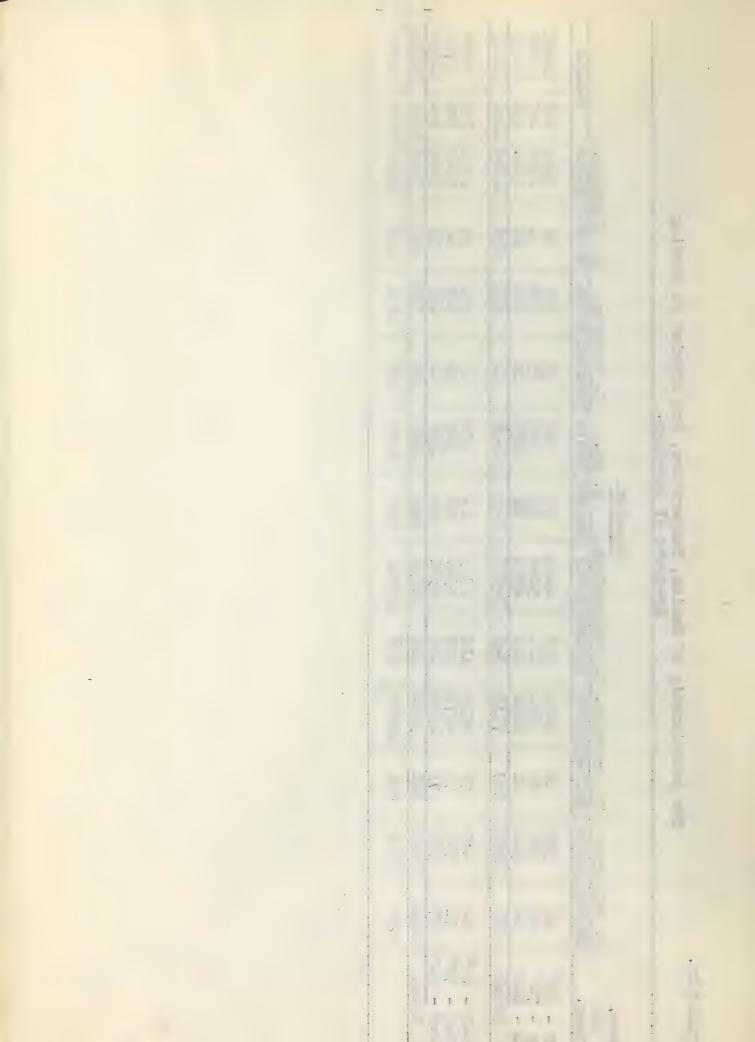
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The Relationship of Medium, Temperature, and Species on Rooting Response of Cuttings.

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		Totals	10682 13369	816	9096 9782 11954 30832 69000	
		To	363 467 450	1280	366 378 460 1204 2484	
	exnosa.	Roots	2561 2703 4800	11064	2441 1941 3472 7854 18918	
	1	Rooted	m9m 200	222	59 62 79 200 422	
	- 14	Roots	251 276 406	933	141 365 233 739 1672	
		Rooted	222		22 40 21 83 164	
	Sp. 0	Roots	525 543 1496		370 309 1350 2029 4597	
pecies	R.	Rooted	WW	149	25 23 90 149 298	
Spe	s. V. tribolum. C. stolonifera	Roots	4466 5268 4774	10	3725 4566 4562 12853 27361	
		Rooted	121	349	112 109 108 329 678	
		Roots	1476 3581 2081	13	2019 1878 1599 5496 12634	
		Rooted	8888		73 75 75 221 468	
		Roots	399 999 560	N	400 723 738 1861 3818	
	S. vulgari	Rooted	61 107 64	232	64 71 87 222 454	
Med	and	Temp.	8 - 60° 8 - 70° 8 - 0ut	Ho	S+P - 60° S+P - 70° S+P - 0ut	



as these two factors are concerned sand has proved the better medium to use. Likewise, medium has not interacted with species or dates to significantly influence the number of cuttings rooted (Table IX). Thus, from the point of view of cuttings rooted, sand has been the more effective medium for each of the individual factors, excepting temperature. However, seldom would cuttings be rooted in sand and be unaffected by more than one of the variables being tested. Usually, two or more are involved at one time. For this reason it would be of more value to state the better medium in relation to the other factors, insofar as possible. Further reference to Tables IX and X indicates that one or two interactions between medium and some of the other factors are significant. Medium and its interactions with the other factors is not the same for the number of roots produced as for the number of cuttings rooted. In Table IX it will be noticed that medium interacts with species and dates to give significant differences. Apparently, these interactions have not affected the number of cuttings rooted. In the relationship of medium to the significant first order interactions with species and dates, it is found that the variance for medium is not sufficiently greater than the variance for either interaction to make the media differences important for different species or dates. Also, none of the higher interactions, including med. x dates, appear significant so that an examination of the med. x dates interaction will indicate the medium and dates relationships. This is shown in Table XII.

It is apparent that, in relation to dates, sand is better than sand-and-peat, but it should be noticed that the advantage of sand over sand-and-peat is not proportionately the same at all seasons. This latter situation accounts for the significant interaction. Sand is slightly more advantageous at the first two dates.

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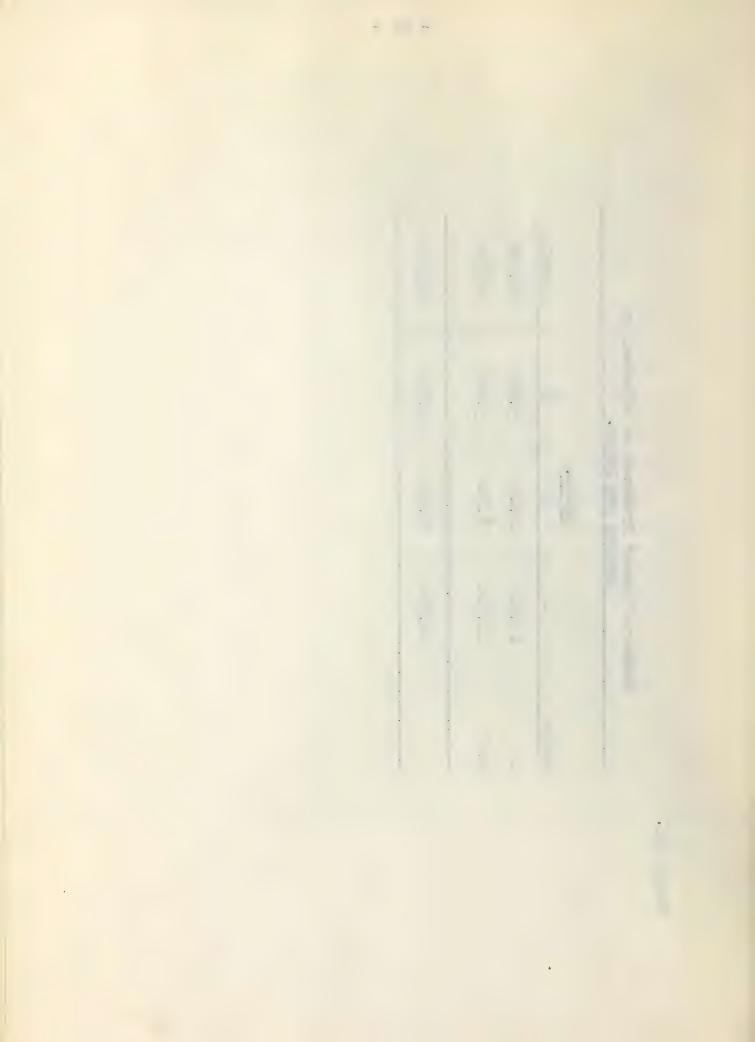
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TABLE XII.

by		Total	38,168	30,832	000.69
as Affected		2	5,971	5,200	171,11
Noots Produced as	Dates.	2	17,275	13,720	30,995
Number of Roots Produced as Affected by Medium and Dates.		7	14,922	11,912	26,834
N		Media	Ŋ	A+0	



Medium and Species Interaction.

The previously mentioned significant med. x spec. interaction affecting the number of roots produced will now be discussed. mean square for the med. x spec. interaction does not significantly exceed the mean square for the med. x spec. x concen. interaction enough to warrant studying the med. x spec. interaction separately. The large variance for med. x spec. x concen. suggests that concentrations influence the med. x spec. results, so that, before conclusions can be drawn regarding the better medium for definite species it will be necessary to consider the influence of concentrations. In addition, the med. x spec. x concen. x cuts interaction shows a large, significant mean square, which would indicate that cuts may have, in some way, influenced the results for the med. x spec. x concen. combination. Statistically, the second order interaction, med. x spec. x concen., is not sufficiently greater than the third order interaction, med. x spec. x concen. x cuts to be significant. Thus, all conclusions, regarding medium by species, must be stated in relation to concentration and cuts. These data are shown in Table XIII.

The better combination of medium, concentration, and basal cut, for the different species, is not readily discernable in Table XIII. However, S. vulgaris responded more readily to a concentration of 40 B.T.I. units and with the basal cut at the node. Within this set of conditions, the differences between the two media are small, but with an indication that the advantage may be in favor of sand. In V. trilobum, sand at 20 B.T.I. units and cut below the node would seem to be the better combination of variables for maximum production of roots. A concentration of 40 B.T.I. units, and cut below the node is the more advantageous combination

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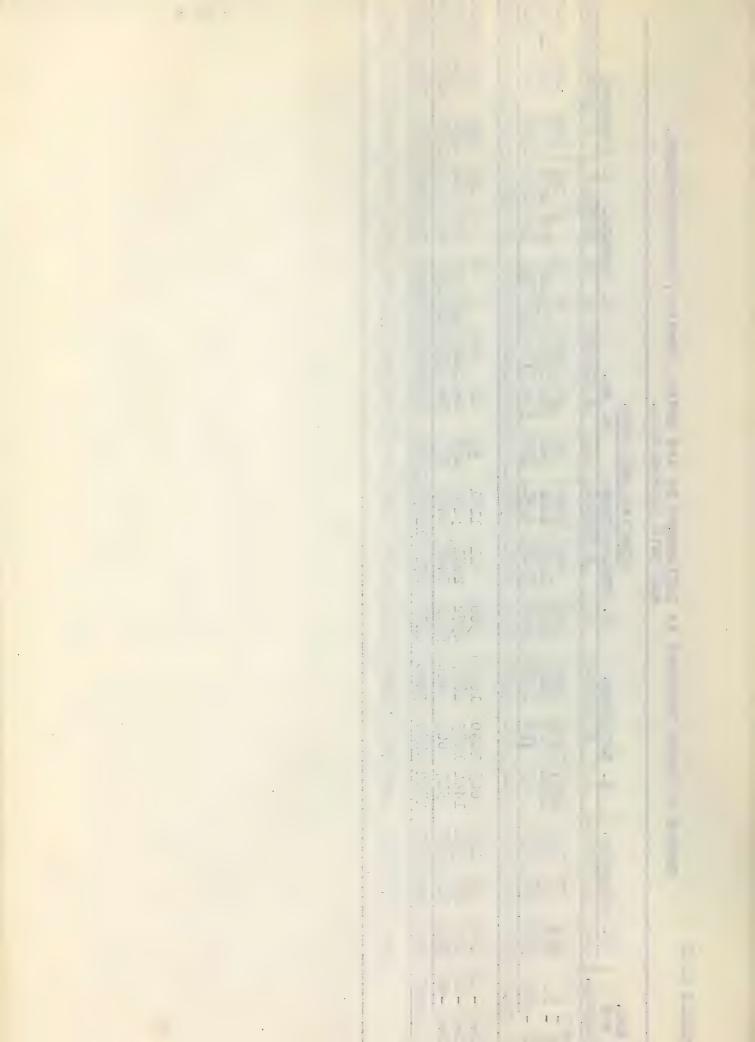
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Influenced by the Media, Species, Concentrations, and Cuts Interaction. Number of Roots Produced as

TABLE XIII.

Med								Speci	0	and Cuts	ts.								
and	S	0.0	404	V.	trilc	ilobum.	C.	stolonif	lifera		R. sp	-	ບໍ	COL	cornuta.	S.	flexuosa	1088	
Concen.	A	В	Total	A	В	Total	A	В	Total	A	20	Tota1	A	B	Tota.	A	B	Total	Total
NN N	上で 50 200 200 200 200 200 200 200 200 200 2	24 074 074	192 693	1097 1476 629	1509	2606 3203 1329	3077 3028	3145 3363	1895 6222 6391	546 570	587	1330	175	202	5735	2951 2769	280 2177 2722	5128 5445 5491	5247 16954 15967
Total	1048	606	1957	3202	3936	7138	7032	7476	14508	1167	1401	2568	412	521	933	5885	5179	11064	38168
S+P - 20 S+P - 40 Total	132 295 565 992 2040	287 287 527 869 1178	187 582 1092 1861 3818	603 1442 317 2362 5564	1090 1435 609 3134 7070	1693 2877 926 5496 12634	560 2572 3001 6133 13165	621 2631 3468 6720 14196	1181 5203 6469 12853 27361	658 373 1130 2297	21 464 414 899 2300	80 787 2029 4597	110 222 339 751	0 203 203 400 400	307 432 739 672	203 350 430 993 878	202 1,886 1,773 3861 9040	405 3246 4203 7854 18918	3546 13377 13909 30832 69000



for <u>C. stolonifera</u>, but, as to the better medium there is little choice. The slight differences between media are in favor of sandand-peat for 40 B.T.I. and at cut B. For <u>S. flexuosa</u> the greatest response has been in sand, at a concentration of 20 B.T.I. units and cut at the node. With the remaining two species, <u>R. sp.</u> and <u>C. cornuta</u>, the differences exhibited are less convincing and further tests would seem necessary before drawing definite conclusions.

Interaction of Medium, Species, Concentrations, and Dates, on the Number of Cuttings Rooted.

The analysis of variance on the number of cuttings rooted (Table IX) showed that there were strong interactions between medium, species, concentrations and dates. It will be remembered from the previous discussion that medium was significant over the small med. x spec. interaction. Furthermore, it will be observed that dates interacts with med. x spec. in a significant way in the med. x spec. x dates interaction, and, in addition, concentrations influences med. x spec. x dates sufficiently to cause a high mean square for med. x spec. x concen. x dates. This significant interaction indicates that the response to different media is not independent of species, concentrations, or dates, or the circumstances which have made one or other of the dates favorable or unfavorable at particular concentrations, or the conditions which have caused any combination of dates and concentrations to be favorable or unfavorable with individual species. The relationships of these four factors may be observed from the data in Table .VIX

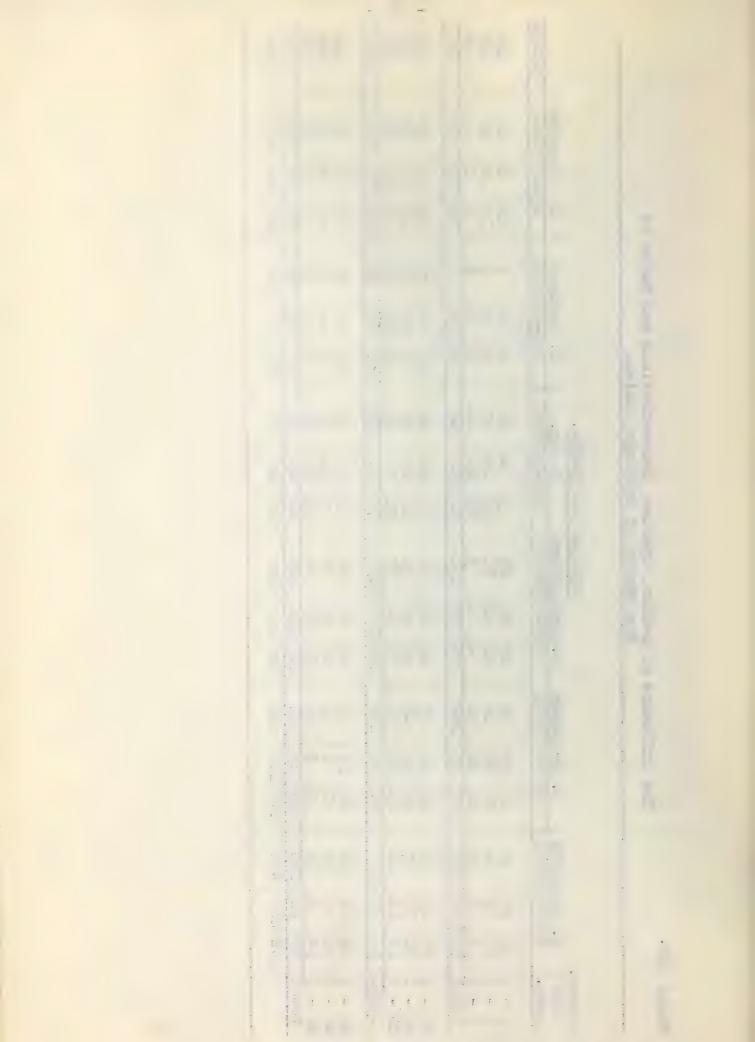
One or two facts stand out in this table. Firstly, the response of S. vulgaris, R. sp., and C. cornuta to the two forms of media do not differ greatly enough at the better combinations

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TABLE XIV.

The Influence of Medium, Species, Concentrations and Dates on the Number of Cuttings Rooted.

	Totals	252 239	587	N	237	3	S	282	10	2484
(Total	226	52	87	300	192	88	12	178	422
Ę	S+P	128	22	41	34	16		49	87	200
. 0	ממ	100	90	46	141	101		41	16	222
4	Total	100	- -1	28	2 H 800	75	62	19	88	164
	S+P	,000	0	9	120	98	30	10	45	83
5	מכ	100	-1	22	<u>~</u> ∞	57	32	00	45	81
Media.	Total	11 27 26	64	46	673	148		-9 -1 -0		298
	S+P	15		24	17	75		20	42	149
s and	Ø	722			202		27	16	44	149
Specie	Tota	80 20 20 20 20 20 20 20 20 20 20 20 20 20	171		12/2		92	69 64	249	819
+	S+P	86	19	46	346	126	45	340	124	329
2	· 10	41 41 4	92	48	39	152	47	31	125	349
mid 0 [S+P Total	88 49 50 50 50	202		52		63	100		468
	S+P	2000		44	27	91	27	-16-	55	221
	S	4 W U	107		25		36	10	46	247
r G	ota	45 42 10	16		28	177	98	981	180	454
[[S+P	26	46		10		44	4 4	91	222
•	מו		51	WI	100	6		72	∞	232
Concen	Date.		Total	00		Total	1	40 - 40 - 70 - 70 - 70	Total	



of concentrations and dates, to warrant drawing any definite conclusions as to the better medium for each set of conditions. This would require additional statistical analysis. Secondly, for the three remaining species, <u>V. trilobum</u>, <u>C. stolonifera</u>, and <u>S. flexuosa</u>, sand has probably given slightly better results at the more favorable combinations of concentrations and dates.

Cuts.

The question of the better place to make the basal cut has not proven to be one of very great consequences. Examination of Tables IX and X shows the differences between the two types of basal cuts to be insignificant. From the standpoint of the number of cuttings rooted, the place of the basal cut is not affected by, nor does it interact in any way with, the other factors to significantly influence the rooting response. As far as the number of roots produced is concerned the same relationship holds, with one exception. That was where cuts in some way affected the med. X spec. x concen. x cuts interaction (see Table XIII) to make it significant. The effect of cuts in this interaction has been indicated when discussing the medium and species relationships under medium.

Temperature.

The statistical analyses show temperature to be a very important factor in securing satisfactory rooting of cuttings. Besides the individual factor of temperature being significant, it interacted strongly with some of the other variables being studied. A few of the more important temperature effects will be briefly discussed.

The significant results for temperature, both for number of cuttings rooted and roots produced, showed the greatest response

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to occur under outdoor conditions, followed next by 70° F., and lastly by the temperature check (60° F.). However, some of the highly significant temperature interactions make it clear that the above order of temperatures does not hold for all conditions within the experiment. In fact, statistically the mean square for temperature alone is insignificant in relation to the mean squares for the temp. x spec. and the temp. x dates interactions. This means that it is essential to state definite species or dates before it would be possible to give the best temperature. Further study of Tables IX and X show that other important interactions occur among temperature, species, concentrations and dates.

Temperature and Species Relationship.

The first relationship to be given consideration will be that of temperature and species. The variance for the temp. x spec. interaction is far in excess of the 1 per cent point when tested against all higher order interactions containing this combination. The strong temp. x spec. interaction indicates that the response of different species varies greatly with different temperatures. Also, the temp. x spec. relationship is influenced by dates, as is indicated by the significant temp. x spec. x dates interaction. However, the mean square for temp. x spec. is sufficiently greater than the mean square for temp. x spec. x dates to conclude that, even though dates interacts with temperature and species in a significant way, the results for temp. x spec. may be used, from which to draw conclusions. This relationship holds for the number of cuttings rooted as well as the number of roots produced. The effect of temperature and species on rooting is shown by the data in Table XV.

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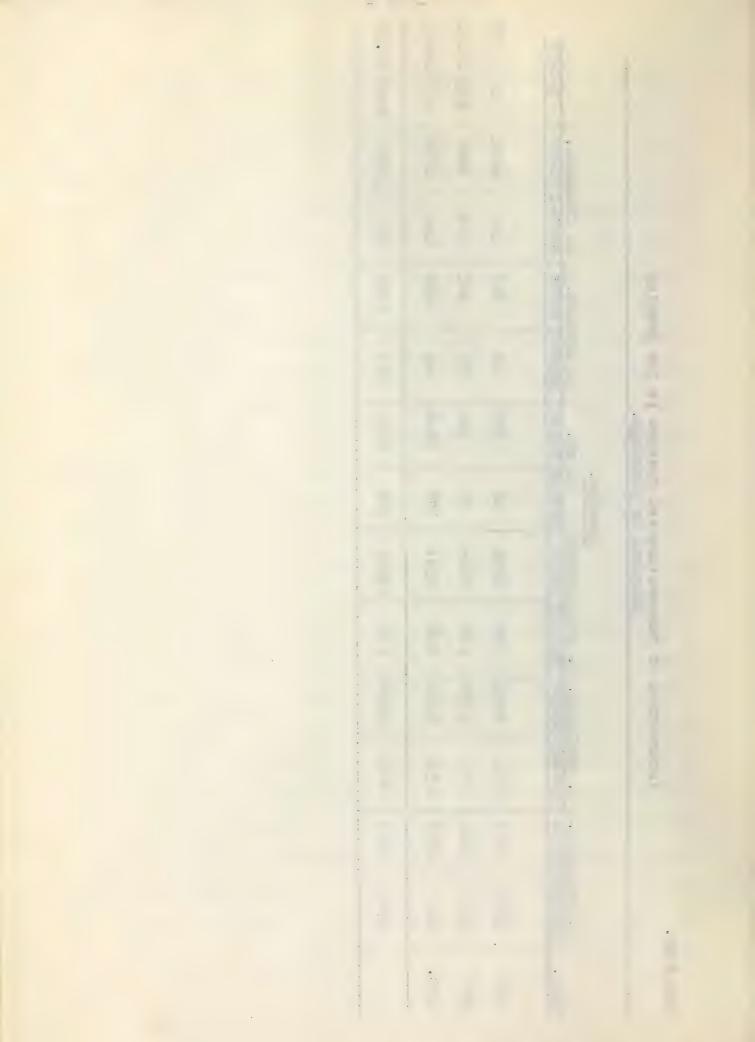
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TABLE XV.

Temperature and Species in the Rooting Response of Cuttings. Interaction of



This table illustrates quite conclusively why the results for the three temperatures, even though significant, should not be cited as the best in the order indicated by the temperature totals. In spite of the fact that the greatest number of cuttings rooted, and the most roots occurred, under outside conditions, the foregoing table indicates this is not the better temperature for all species. The significance of temperature really means that outside was the better general temperature condition for all species tested.

observed. S. vulgaris, V. trilobum, and C. cornuta rooted better, and produced more roots at 70° F. than at the other two conditions. In the case of C. stolonifera the effect of temperature was less marked, but again 70° F. appeared to be slightly more satisfactory. The outside condition was more favorable for S. flexuosa, and for R. sp. was definitely the best. This latter fact was very noticeable in the rooting frames during the course of the experiment.

The significant temp. x spec. x concen. interaction (see

Table IX) will be left for the present and discussed under "species."

Temperature and Date Relationship.

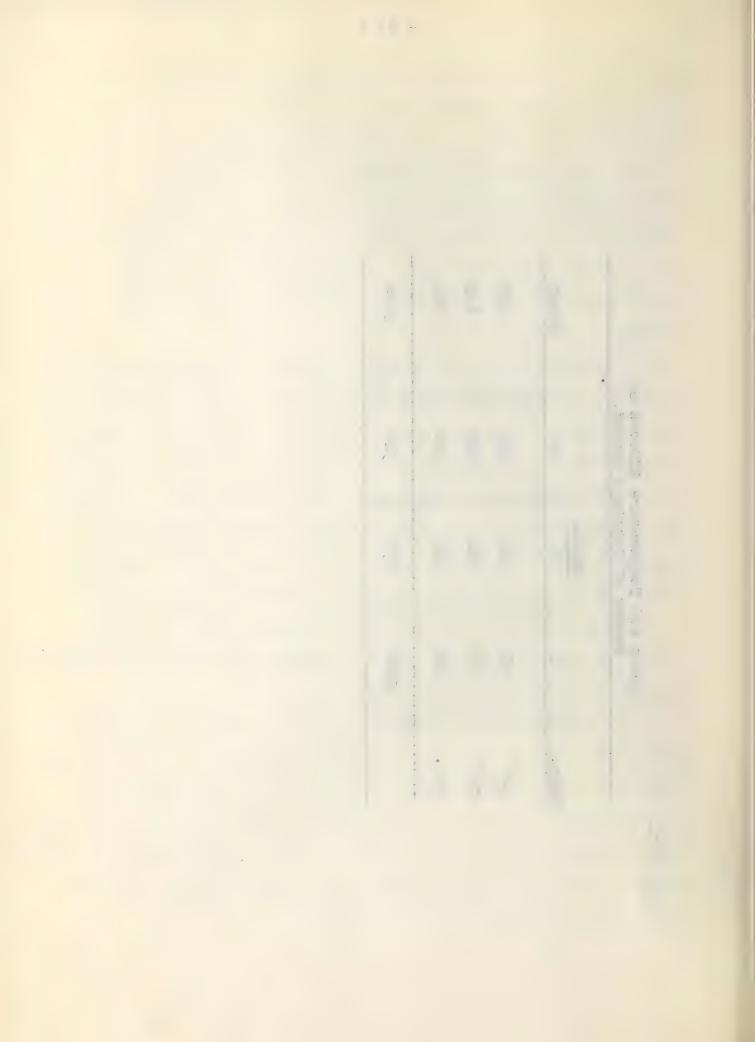
The relationship of temperature to the temp. x dates interaction has already been noted. In addition to the temp. x dates interaction being significant, its mean square is sufficiently greater than the mean square for the significant temp. x spec. x dates interaction to justify a discussion of the temperature by date results separately from species. The following table shows this first order interaction.

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TABLE XVI.

T.	Total		729	845	910	2484
e Effects on	K		1.67	187	204	528
Date and Temperature Effects Number of Cuttings Rooted.	Dates.	2	677	322	323	858
Date and Numbe	1	270	213	336	383	1098
	Temp.	009	3	700	Out.	



This table sets out quite clearly the temperature and date interaction. Outside temperature has given slightly better results at all three dates, but at date 2, 70° F. was just as effective. At the first date 60° F. was better than 70° F., while at date 3 60° F temperature gave the poorest results.

The preceding discussion of temperature and date interactions applies only to the number of cuttings rooted, and not to the quantity of roots produced. In the case of the latter, the temp. x date interaction is not sufficiently greater than the temp. x spec. x date interaction to justify discussing temperature and date effects apart from species. Furthermore, the variance for the temp. x spec. x seas. interaction, both as to cuttings rooted and roots produced, is not statistically significant over the variance for the temp. x spec. x concen. x date interaction. This indicates that the temperature effects are not independent of the other three factors involved, or of the conditions which have made certain species respond better at certain concentrations, or of the conditions which have made certain combinations of species and concentrations more favorable at specific dates. Thus, it would seem more correct to draw conclusions in regard to temperature, only after stating the species, concentration and date. The interrelationship of these four factors can best be obtained from the data in Tables XVII and XVIII.

The only conclusions to be drawn, from these temp. x spec. x concen. x date interaction tables, will be those involving the more desirable combination of temperature, concentrations, and dates for each species. Then, the most favorable combination of these four factors, for root production, would be somewhat as follows:
For S. vulgaris, best results, for rooting and roots produced, have

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Effect of the Interaction of Temperature, Species, Concentration and Date on the Number of Cuttings Rooted.

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	-	otal	100		52		63		192	55	56	67	178	422	-
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	$\dashv \Box$	7	10	10	23		500		75		21		75	173	
	က္ခု	-	<u>~</u>	11	56		30		87	31	31	56	88	10	
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		7	00	0	0	Н	15.1	2	19	M	11	2	19	38	
	0	-1	10	0		0	10	2	28		25	14	2	16	
	•	otal	0/0	49	64		27		148		10		86	298	
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Dat	MI.	N	10	24	27	0	2		37	H	N	14	17	81	
nd		-1	40	10	11	20	5	21	46	0	0	24	33	06	
හ ග	ra.	otal	200		171		000		258		86		249	678	
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		2	4 K	11			29		67		31		16	185	-
	വ	-	97	100	45	31	200	23	82			29		213	
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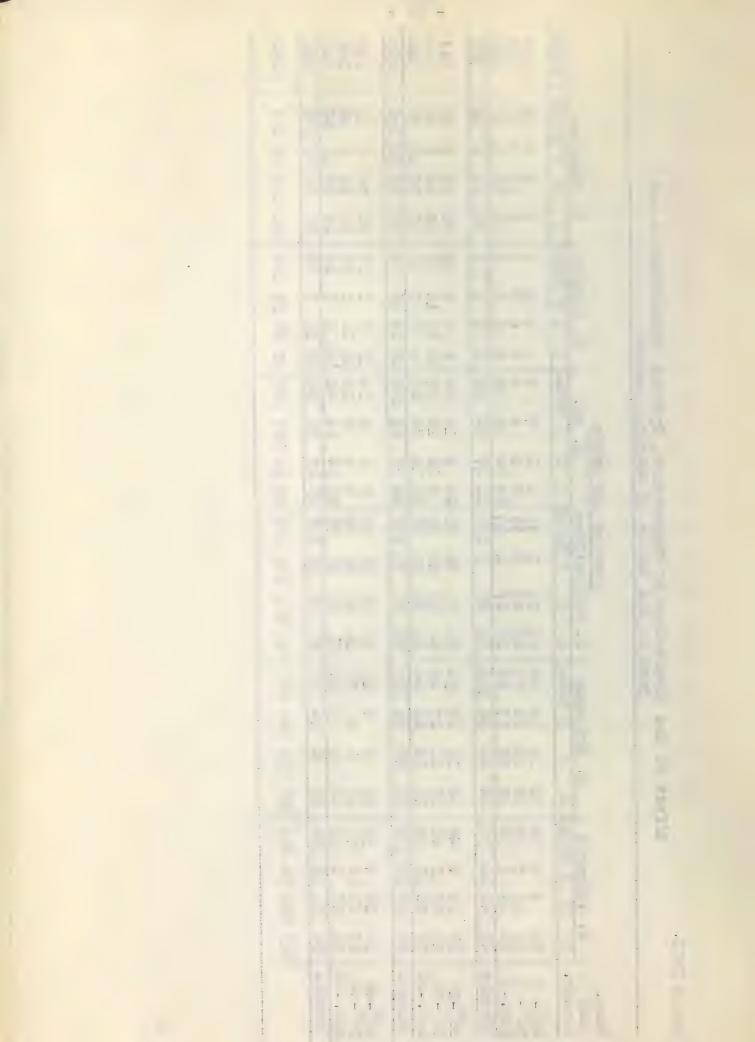


TABLE XVIII.

Effect of the Interaction of Temperature, Species, Concentration, and Date on the Number of Roots Produced.

Temp.									1	Spec	cies	and Dat	es.												
and		s. vul				V. tr	ilob		(C. stol	lonife			R.	sp.			C. cc	ornu	ta		S. f.	lexuo	sa	
Concen	1	2	3 1	Cotal	1	2	3	Total	1	2	3	Total	1	2	3	Total	I	2	3 !	Total	1	2	3	Total	Total
60°- 0 70°- 0 out0	47 68	19 110 73	9 19 1	61 176 142	756	316 656 385	221 637 279	1420	469 552 476		39 16 3	1089	2 0 27	2 3 82	18 12 38	22 15 147	5 0 0	0 0 0	0 0 0	5 0 0	226		0 8 1	244 236 370	3257 3168
Total	148	202	29	379	1805	1357	1137	4299	1497	1521	58	3076	29	87	68	184	5	0	0	5	520	321	9	850	8793
60°-20 70°-20 Out.20	120 122	92 348 313	21 87 43	242 555 478	1064 943 759	477 894 543	306 786 308	1610	1122	2118 1818 2015	553 1040 452	3821	281 147 209	115 161 270		634 1144		57 11	57 78 59	153 313 219	1516	1259 913 1388	180 141 795	1937 3699	10042 109 7 1
Total	371	753	151	1275	2766	1914	1400	6080	3429	5951	2045	11425	637	546	1309	2492	421	70	194	685	3690	3560	1116	0274	30331
	277	400	24 107 1	496 990 678	598 703 641	0 38 5	23 243 4	650	1161 1093 1380	1990 2820 2693	407 963 353	3558 4876 4426	120 0 441	18 76 416	-	1555		7 77	7 5 105	328 420	1163 1500	2282	15 127 421 563	2471 4203	8092 9852 11932 29876
Total	750	1282	132	2164	1942	43	270	2255	3634	7503	1723	12860	561	510	850	1921	687	1.18	11/	982	2724	5197	207	7074	27010
	1269	2237	312	3818	6513	3314	2807	12634	8560	12975	3826	27361	12 2 7	1143	2227	4597	1113	248	311	1672	8152	9078	1688	18918	69000



been at a temperature of 70° F., at date 2, and for a concentration of 40 B.T.I. units. In C. cornuta, the same set of conditions hold, except that date 1 rather than date 2 has been more preferable. Outside conditions at date 1 and 40 B.T.I. units has been the most advantageous for number of roots produced in S. flexuosa, but for number rooted date 1 at 70° F. seems best. In this species, the advantage of a concentration of 40 B.T.I. units over 20 B.T.I. is slight, when the other two factors are optimum. Good results were secured in Rosa sp. at date 3, under outside conditions, and a concentration of either 20 or 40 B.T.I units. C. Stolonifera responded reasonably well over a comparatively wide range of conditions, but better rooting occurred at 40 B.T.l units, date 2, and at 70° F. As for V. trilobum, the first date consistently gave better results when in combination with a concentration of 20 B.T.1. The temperature effects in this latter combination are not clearly evident. although a slight advantage has occurred in favor of 60° F.

Species.

The relationship of species to the other factors being studied will not be considered. The effects of temperature and media on species, and their interactions with species, have been briefly discussed already. It was pointed out previously that cuts is not an important factor, so that only the relationship of species to concentrations and dates remains.

The results for species differences are highly significant in terms of the grouped interactions containing species. Also, it is apparent that the response of different species is not independent of concentrations, nor of dates, as pointed out by the strong interactions of spec. x concen., and spec. x dates. However, statistically it is found that the variance for species is sufficiently greater than the variance for both of the above interactions to

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exceed the 5 per cent point. Similarly, "species" is significant in relation to the temp. x spec. interaction. These facts make it possible to draw conclusions respecting the order in which the different species root most readily, as well as the order of their ability to produce roots. Reference to Table XVII shows C. stolonifera to have rooted most readily, followed in order by V. trilobum, S. vulgaris, S. flexuosa, R. sp., and C. cornuta. Table XIX reveals the fact that C. stolonifera has produced the greatest number of roots, followed by S. flexuosa, V. trilobum, S. vulgaris, R. sp., and C. cornuta.

Species and Concentration Relationship.

The question of the greatest number rooted, or the most roots produced, does not present the whole picture. For instance, the highly significant first order interactions, of spec. x concen., and spec. x dates, indicate that all species have not reacted to the same extent within any one concentration, or any one date. It will be necessary to consider these interactions in order to arrive at more specific conclusions as to the concentrations and dates that will give maximum results for each species. Statistically, both first order interactions are significantly greater than the strong spec. x concen. x dates interaction, and this means that the better concentration and better date for each species may be obtained from tables for these first order interactions. First, the effect of concentrations on species will be examined. (see Table XIX).

From the standpoint of number of cuttings rooted and the number of roots produced, S. vulgaris and C. cornuta responded better at the concentration of 40 B.T.I units. In R. sp. the treatment involving 20 B.T.I units was the most favorable. The

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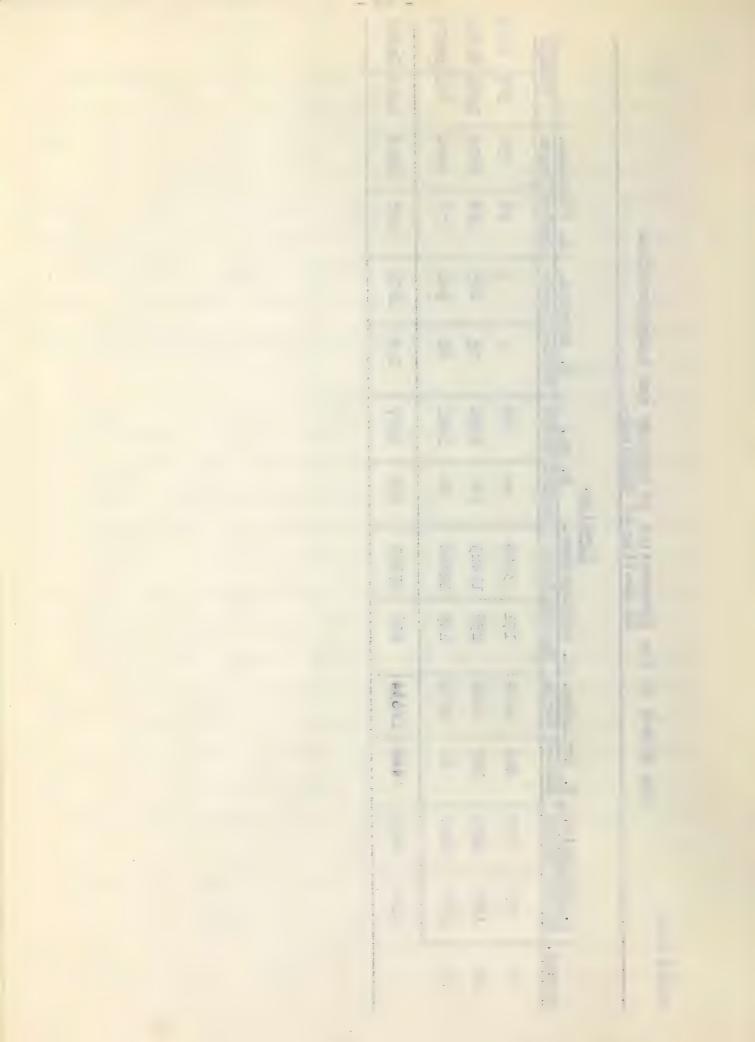
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TABLE XIX.

The Effect of the Interaction of Species and Concentrations on Rooting of Cuttings.



better concentration is less evident in the remaining 3 species.

Slightly more rooting occurred in C. stolonifera and S. flexuosa
when the treatment was 20 B.T.l units, while greater root production resulted from treatment at 40 B.T.L units. However, for
C. stolonifera and S. flexuosa the differences between the results
for the two higher concentrations (20 and 40 B.T.L.) are rather
small for the number of cuttings rooted, but large for number of
roots produced. Possibly as point in between these two concentrations would give a maximum rooting response. In V. trilobum
20 B.T.L units appears to be near the upper limit for concentrations.

The necessity of the proper concentration to obtain maximum rooting response was well illustrated in the 1938 experiment. The range of effective concentration was much wider in C. stolonifera than in the other species. The opposite tendency was noted in R. sp., V. trilobum and S. flexuosa. In S. flexuosa, some burning of the basal portion of the cutting occurred at the 40 B.T.I. concentration. This is illustrated in Fig. III. The effect of differences in concentration were even more marked in V. trilobum. Here, the total number of cuttings rooted was considerably greater at zero than at either of the higher concentrations, although more roots were produced at 20 B.T.I. units. This advantage in favor of the check may be partially due to both higher concentrations, especially 40 B.T.I., being too strong and causing considerable burning of the treated ends of the cuttings. This effect is illustrated in Fig. IV. A number of preliminary trials suggests that 10 B.T.I. units is probably a better concentration for V. tri-These same trials indicated that 20 B.T.I. units was approximately the upper limit of concentration for S. flexuosa.

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Fig. III. Spiraea flexuosa - showing response to different concentrations of Hormodin A. From the left the concentrations applied were 0, 10, 20 and 40 B.T.I. units, respectively. At 40 B.T.I. units some burning of base of the stem occurred.

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Fig. IV. Viburnum trilobum - showing rooting response at varying concentrations of Hormodin A. Beginning from right to left the concentrations used were 0, 10, 20 and 40 B.T.I. units, respectively. Notice burning of the base of the stem at 40 B.T.I.



Species and Date Effects on Rooting.

The next question considered is the best date of the year to gather the various species. It will be recalled, that, in the discussion of species and concentrations, the mean square for spec. x dates was shown to be significantly greater than the mean square for the spec. x concen. x dates interaction. Examination of the strong spec. x dates interaction (Table XX) enables certain conclusions to be drawn regarding the best dates to gather the different species.

The results for V. trilobum, and C. cornuta show that the better time to gather these two species is date 1, i.e. somewhere about June 20. Of course, this will depend upon the maturity of the cutting material, but the indications are that cuttings of these two species should be taken as soon as the wood is sufficiently mature. In the Rosa sp. the reverse seems to be the case. The indications are that better rooting response may be expected with cuttings taken around date 3 (early August). The remaining three species, S. vulgaris, C. stolonifera, and S. flexuosa, from the standpoint of number rooted and number of roots produced, show different responses at various dates. In every case the greatest number rooted when gathered at date 1, while more roots were produced in all three species at date 2. This latter fact would seem to suggest that there may be a period between dates 1 and 2 where a balance, between number rooted and roots produced, might be reached such that the largest number would root and produce the greatest number of roots.

It should be borne in mind that the foregoing conclusions from the data in Tables XIX and XX apply to the general or overall effect of the different concentrations and dates on species.

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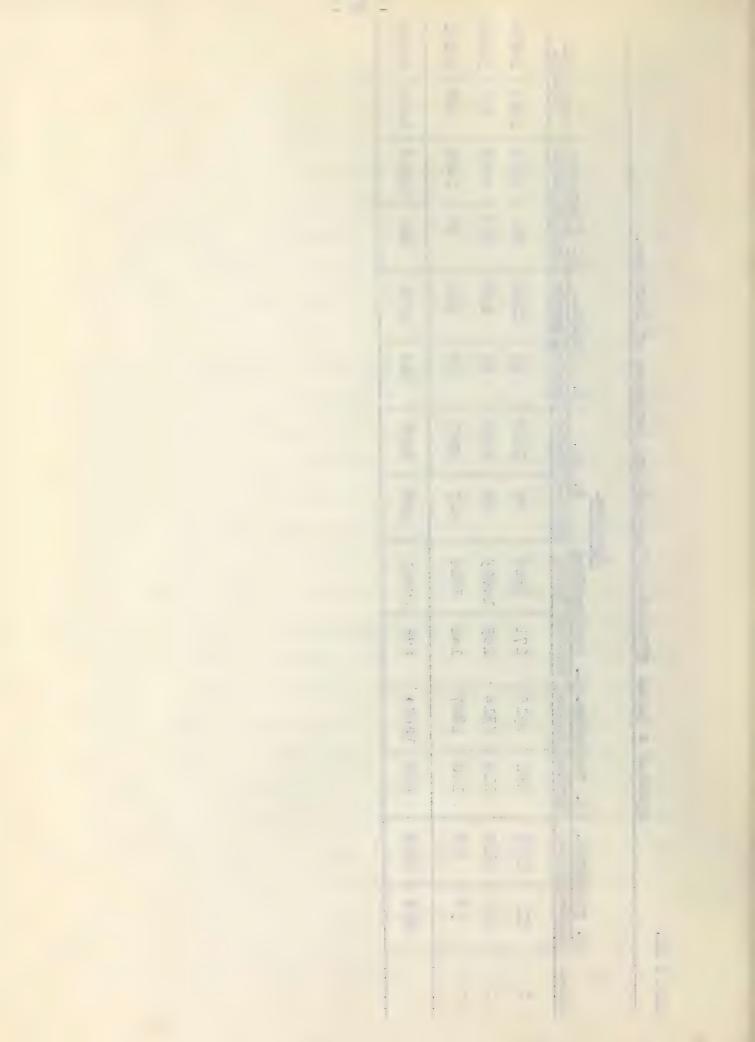
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Species x Dates Effects on the Rooting Response of Cuttings.

		Totals	26834	30995	11171	00069
	«Webson	Tot	1098	858	528	2484
	xuosa.	Roots	8152	9078	1688	18918
	S. flexuosa.	Rooted Roots	201	173	48	422
	nuta.	Roots	1113	248	311	1672
	C. cornuta.	Rooted Roots	91	38	35	164
	R. Sp.	Roots	1227	1143	2227	4597
Species.	R.	Rooted	06	81	127	298
Spe	stolonifera	Roots	8560	14975	3826	27361
		Rooted	271	265	142	678
	trilobum. C.	Roots	6513	3314	2807	12634
	V. tri	Rooted	232	116	120	468
	vulgaris.	Roots	1269	2237	312	3818
	S. vul	Rooted	213	185	56	454
		Dates	Н	2	W	



These conclusions do not give any suggestions as to the smaller fluctuations brought about by the interaction of species, temperature, concentrations and dates. The significant interactions of these four factors has been discussed under temperature, in connection with Tables XVII and XVIII.

Concentrations and Dates.

The effects of concentrations and dates will be discussed together. It will be observed from the analyses of variance tables, for cuttings rooted and roots produced, that the results for each variable are highly significant. For the number of cuttings rooted, the differences obtained at the various dates (see totals table XX) are significantly greater than all first order interactions containing dates, and this means, in general, that the first date is the best time to gather the cuttings. As regards the number of roots produced, the mean square for dates is significant over the mean square of the concen. x dates interaction, but in relation to spec. x dates it is not. This indicates, that for all concentrations, date I would be expected, in general, to give better results, but, for all species no one date is consistently the best.

The variance for concentrations is barely significant over the variance for spec. x concen. Such a relationship would suggest, that insofar as species are concerned, one generally best concentration cannot be stated without first specifying the species. However, concentrations is significant in terms of the concen. x date interaction. From this it may be concluded that for all dates a concentration of 20 B.T.I. units was the better (see Tables IX and XIX). Also, the analysis, of the number of roots produced, shows that concentrations bear the same relation-

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ship to species as has already been pointed out for dates and species.

Although the above generalized relationships, as regards concentrations, and to dates, were apparent, they do not tell the whole story. Some higher, significant interactions, containing concentrations and dates, indicate that for certain sets of conditions, within the experiment, these general conclusions may not be valid. Thus, to secure more practical information on the proper conditions for maximum rooting, it is necessary to base any further conclusion on the higher order interactions. attention will be directed to the concentration and dates reaction. In Tables IX and X the mean square for concen. x dates is not sufficiently greater than the mean square for spec. x concen. x dates to be significant. The variance for the spec. x concen. x dates interaction in relation to the variances of higher interactions containing this combination is highly significant. Because of these two facts, it would be more advisable to draw conclusions regarding combinations of concentrations and dates, only after specifying the species. The significant influence of species on the concen. x dates interaction is shown in Table XXI.

A few conclusions as to the better combination of concentration and date for each species may be obtained from this table. Undoubtedly, the better set of conditions for <u>C. cornuta</u> was date 1 and a concentration of 40 B.T.I. units. On the other hand, Rosa sp. was most strongly influenced by a concentration of 20 B.T.I. at date 3. Best results were secured in <u>V. trilobum</u> at 20 B.T.I. units and date 1, while in <u>C. stolonifera</u> greatest response occurred at date 2, and a concentration of 40 B.T.I. units. For the two remaining species, <u>S. vulgaris</u>, and <u>S. flexuosa</u>, the more favorable set of conditions is less evident. In both cases,

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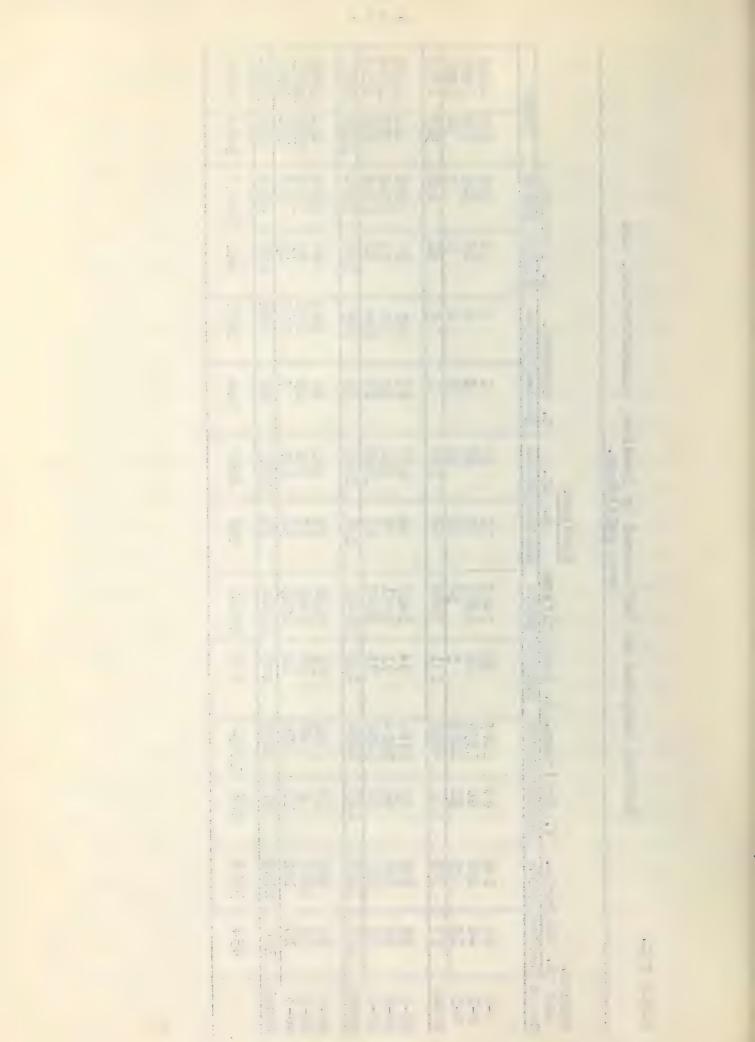
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Rooting Response as Influenced by Species, Concentration, and Date Relations.

TABLE XXI.

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	Tota			96	587	0	337	-	1035	24	282	5	N	2484
flexuosa.	Roots	(2200		850	69	3560	H	8374	93	5197	56	9694	18918
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cornuta.	Roots	ı	vc	00	2		70	194	685	∞	174	H	982	1672
C. corr	Rooted	•	-10	00	r-1		19		75	62	19	7	88	164
Sp.	Roots		200	89	184	N	546	0	2492	9	510	5	1921	4597
Species R.	Rooted		11	56	64		37		148		17		98	298
4	Roots	6	1447 1031	10	3076	42	5951	04	11425	63	7503	72	12860	27361
stol	Rooted		000	150	171		16	73	258		93		249	678
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0 0	Dates		100		Total	0	20-2	0	Total	40-1	40-2	40-5	Total	



the greatest number rooted at date 1 and 40 B.T.I., whereas more roots were produced at date 2 and 40 B.T.I. units.

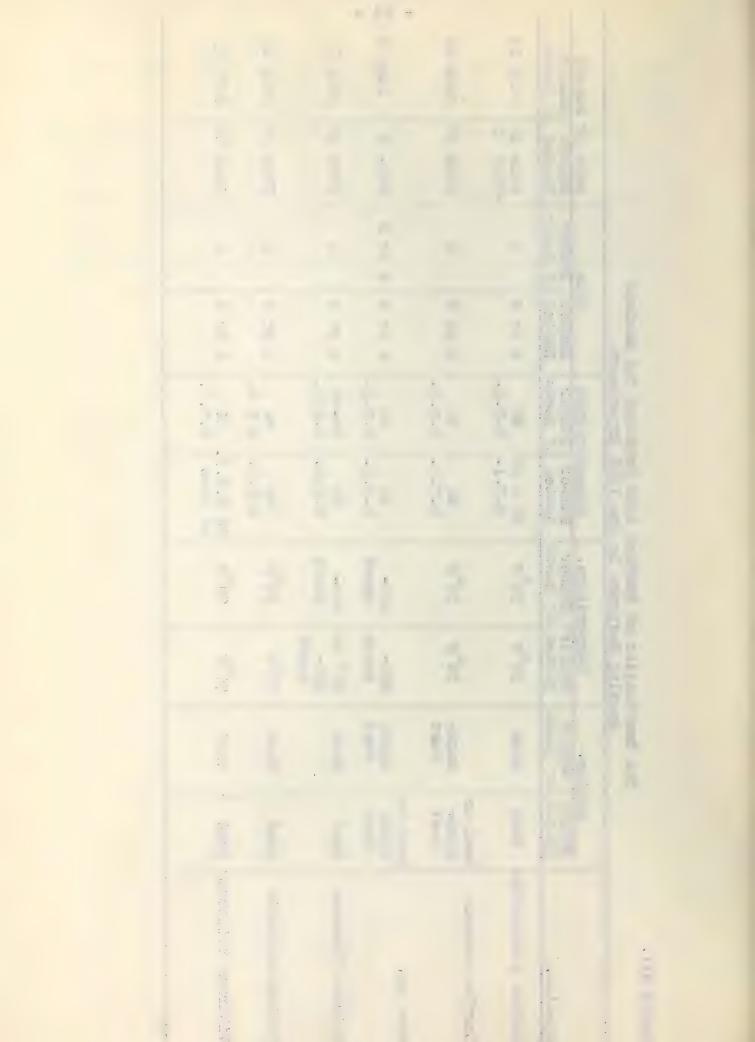
From the practical viewpoint, this discussion of results may be summarized in a manner similar to that for the 1937 investigation.

A summary table, based upon the analyses and results, is presented to show the particular combination of variables, which, in this experiment resulted in maximum rooting response (see Table XXII).

TABLE XII.

The Combination of Factors that Produced the Maximum Rooting Response in the 1938 Experiment.

		HOO	ागल प्रवा	TIT ASTION	ROOTING RESPONSE IN THE 1920 EXPERIMENT.	EXPER INC)II C •			
	Med	Medium.	Tempe	Temperature.	Concent	Concentration	Cu	Cuts	Date of Season	Season
Species.	Number Rooted	Number of Roots	Number	Number of Roots	Number Rooted	Number of Roots	Number Rooted	Number of Roots	Number	Number of Roots
Cornus stolonifers	Sand	Sand	70°F	70°F	20 - 40 B.T.I.	40 B.T.I.	A or B	д	June 20 July 15	July 15
Corylus cornuta	Sand or Sand+ Peat	Sand+ Peat	70°F	100F	40 B.T.I.	40 B.T.I.	A or B	щ	June 20	June 20
Rosa sp.	Sand or Sand+ Peat	Sand+ Peat	Out-	Out-	20 B.T.I.	20 B.T.I.	A or B	A or B	Aug. 8	94 - 94 - 94 - 94 - 94 - 94 - 94 - 94 -
Spiraea flexuosa	Sand	sand	70° or Out-	Out- doors	20 B.T.I.	20-40 B.T.I.	A or B	4	June 20	July 15
Syringa vulgaris	Sand	Sand	70°F	70°F	40 B.T.I.	40 B.T.I.	A or B	4	June 20	July 15
Viburnum trilobum	Sand	Sand	70°F	70°F	20 B.T.I.	20 B.T.I.	A or B	щ	June 20	July 15



GENERAL DISCUSSION.

The most important feature of these experiments, as shown by the analyses of the results, is the significant interactions that occur among five of the seven variables studied, namely, temperature, species, treatment, concentrations, and the time of season when the cuttings are taken. The remaining two variables, medium and basal cut, in most cases did not materially influence the responses obtained.

Numerous workers have shown that the rooting response of different species of plants varies with the phytohormone used, as some acids are more stimulative than others. They have shown, also, that the response varies with the concentration of acid used, with the temperature at which the cuttings rooted, and with the time of season when the cuttings are gathered.

The stage of maturity of the cuttings is an important factor in ensuring good rooting, and is, of course, directly correlated with the time of season. The present investigation shows, further, that all species of plants do not react alike at the same stage of maturity. Also, it shows that different temperatures bring about different responses at various times of the season. Likewise, temperature is found strongly to affect the action of the different phytohormones tested, and the concentration which encourages maximum rooting varies with the dates.

This interaction of one variable with another, as indicated in the foregoing paragraph, is to be expected. However, the influence of one factor on another, for example, temperature on a particular species, does not stop at the point suggested above. In fact, it becomes increasingly apparent that the rooting response produced by the influence of any one of the factors - temperature,

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treatment, concentration, or date gathered - on individual species. is not independent of the other factors. Statistical proof is given to show that the effect of any one of the above factors on the response produced by two other factors is usually too great to enable any conclusions to be drawn regarding the relationship of the two. For example, it is known that different species require specific concentrations, but, in this study it is found that both temperature, and time of gathering the cuttings, influence the concentration and species relationship sufficiently so that the best concentration for any species should not be stated without specifying the temperature used in the rooting process, and the stage of maturity at which the cuttings are taken. Therefore, simple recommendations, that a definite temperature is best for individual species, or that a certain acid is more effective for certain species or that a specific concentration is best for a definite species, cannot justifiably be made without knowing the other variables involved. Such recommendations occasionally may, but usually do not, present the full set of conditions necessary to encourage maximum rooting response.

Since the factors of temperature, sensitivity of species, treatments, concentrations, and date of gathering the cuttings are unavoidably present in all experiments of the nature of the ones being reported, and in the commercial propagation of greenwood cuttings, it becomes imperative that further information regarding the complex interaction of these factors should be obtained. Then, more specific recommendations for maximum rooting response could safely be made. Probably many of the unsatisfactory results now secured with commercial preparations of phytohormones may be due in part to insufficient attention being given, in the directions,

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and also in the carrying out of these directions by the propagator, to certain of the variables, namely, temperature and maturity of the wood. It would seem highly desirable to investigate this matter further.

The rooting response, as affected by the two types of media used in these investigations, sand and a sand-and-peat mixture, is about equal. In fact, the differences are not consistent enough, in any of the species tried, to safely pronounce either medium as the better. The results in the sand medium, on the whole, are probably slightly better than those in the mixture. According to Chadwick (3), Hitchcock (8), and Laurie and Chadwick (14), a mixture of sand and peat moss is superior to other media in the majority of plants studied. They found that sand gives good results in a few plants but that the response was much less consistent than in the mixture. Hitchcock (8) attributed much of the success of the sand-and-peat mixture to the presence of what he termed "growth promoting materials" in the peat moss. He based this assumption on the fact that root growth proceeded more quickly in the mixture than in sand, and upon the fact that the form of growth was often quite different. If growth-promoting, or at least stimulative, substances are actually present in the peat moss, it is readily understood, with the present knowledge of phytohormones, how they could have stimulated the cuttings to root more quickly and profusely, and how the sand-and-peat mixture would then be acclaimed the best medium. In the present study, the cuttings received the root-promoting substances before they were placed in the rooting medium. This treatment would then do away with the necessity of receiving any stimulative action from the rooting medium, so that sand would then be as effective as the

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mixture.

Another point that may account for the disagreement between the results of this study, and those of Chadwick (3), Hitchcock (8), and Zimmerman and Hitchcock (37), is that the peat moss and sand-and-peat mixture, used by them, was decidedly acid in reaction. The mixture used in these investigations was nearly neutral in reaction.

In this report, the type of medium is not a factor of great importance. However, one or two secondary considerations may be of sufficient concern to determine which type of medium should be used. It is found that cuttings rooted in sand consistently possess larger, and generally less branched, roots than those in sand-and-peat. It is not known whether this difference in size is of anatomical or physiological importance to the plant. The analyses show that more roots are produced in the sand than in the mixture. The production of more and larger roots may, in some cases, be an important feature in favor of sand. When the cuttings were removed from the sand-and-peat the ends of many of the longer roots broke off. This was probably due to a tendency of the mixture to pack more firmly than sand. In the fine-rooted species especially, this breakage was very noticeable.

The relationship of temperature and media may be a deciding factor as to which type of medium to use. Chadwick (3) showed that sand-and-peat has a 3° to 4° F. higher temperature than sand, and furthermore, the temperature remains more constant in the mixture. The same situation was observed in uncontrolled green-house temperatures in the present investigations. Zimmerman and Hitchcock (37) showed that the time of rooting, in Ilex verticillata, was reduced seven days by an increase of 4.5° F. from 68° to 72.5°F. Because of the tendency for sand-and-peat to raise the

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temperature several degrees, the use of the mixture may be advisable when there are no facilities to apply bottom heat.

In the present studies there is no consistent evidence to support the idea that sand-and-peat is more favorable than sand, either in the greenhouse in the check frames, or under outdoor conditions. In fact, better rooting generally occurred in the sand. The fact that the cuttings were rooted during the summer months, when the average daily mean temperature is highest, may have altered the tendency found by Zimmerman and Hitchcock (37).

Before the application of phytohormones came into use as an aid in the vegetative propagation of plants by greenwood cuttings, the place of the basal cut had an important bearing on the success of the rooting process. In these investigations the place of the basal cut proved to be of little consequence in cuttings treated with phytohormones. In no cases did the place of the basal cut significantly affect the number of cuttings rooted. As pointed out by Chadwick (10), and Priestly and Swingle (24), normally the ability of a cutting to root depends upon three closely associated factors: (1) the presence of root initials, (2) healing of the wound, and (3) a sufficient food supply. In untreated cuttings the production and number of roots, their placement on the cutting, etc., depends largely upon the presence of pre-existing root initials. Species of plants differ in respect to where the most root primordia are situated. The common belief is that, in most species, they occur mainly in the region of the node, but in some are present in the internodes. The most effective place of the basal cut would then appear to be governed by the distribution of the root primordia. In the hormone-treated cuttings the existence of pre-formed root initials does not seem to be so essential to en-

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sure good rooting. Howard (11) has pointed out that phytohormones most likely act as root determiners, and are able to cause the meristem in the cambial region to send out roots rather than shoots. Thus, a treated cutting may be induced to send out roots from points where no pre-existing primordia were present. This effect of the growth-promoting substances was very noticeable on most treated cuttings. Hence, it becomes clear why the place of the basal cut, under such circumstances, could not influence the rooting process.

There is some indication that the place of the basal cut may be important in phytohormone treated cuttings in respect to the number of roots produced. For instance, untreated cuttings of Viburnum trilobum generally produce roots from the nodes. Because of this, better rooting is usually obtained when the basal cut is made below the node, since the cut at the node may remove some of the root initials. In treated cuttings, roots arise in the internodes as well as at the nodes, and, in the light of what has been said regarding the root initiative effect of the phytohormones, it might be supposed that the place of the basal cut would have no effect on the root production. However, it is still found, after treatment, that more roots are produced per cutting, when the cut is made below the node. Just the reverse situation appeared in Spiraea flexuosa. In most of the species studied the differences in number of roots produced, whether cut at the node or below, are not large enough or sufficiently consistent to make any definite conclusions possible. In any case, from what has been said in this and the preceding paragraph it is evident that the place of the basal cut is a minor consideration in the propagation of plants, by means of the greenwood cutting treated with phytohormones.

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In the 1937 experiment, indole-acetic and indole-butyric acids are definitely superior to indole-propionic and naphthalene-acetic acids. The results do not show any very great advantage in favor of indole-acetic over indole-butyric, or vice versa. However, when in combination with the optimum conditions of temperature and concentration the indole-butyric acid is usually slightly better than indole-acetic acid. On the other hand, the indole-acetic acid is effective over a wider range of concentrations.

Naphthalene-acetic acid gives the least stimulation of all. This fact rather contradicts reports by Hitchcock and Zimmerman (30, 31), in which naphthalene-acetic acid was equally as effective as indole-butyric acid. They claim also that naphthalene-acetic acid produces stimulation at lower concentrations than indolebutyric, and indole-acetic acids. Since, the same strengths of solutions were used in the four acids, the poor results for naphthalene-acetic acid may have been caused by too strong concentrations. However, this did not appear to have been the case. In the first place, there was no marked evidence of burning of the treated portion of the stem, which so often accompanies too strong solutions. Secondly, in Lonicera tatarica and Cotoneaster acutifolia there was an increase in cuttings rooted, as well as number of roots produced, for an increase in concentration from 0 to 40 mg., but in Syringa villosa a decrease in rooting response occurred for an increase in concentration up to 40 mg.. So far as can be stated at this time, the differences in stimulative effects do not appear to explain the superiority of indole-butyric and indoleacetic acids over naphthalene-acetic acid.

There is a strong indication that temperature may have had some effect on the stimulative action of naphthalene-acetic acid.

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The 60° F. temperature resulted in greater rooting than did 70° F., while just the reverse was true in the other three acids. These results would seem to indicate that naphthalene-acetic acid was relatively more effective at lower temperatures than indole-butyric, indole-acetic, and indole-propionic acids. This differential response to temperature may be one reason for the favorable results obtained for this acid by other workers. Further experimentation will be necessary to clarify this point.

Satisfactory rooting was obtained in all the species tested, except one. When the combination of temperature, treatment, concentration, and maturity of the wood was the most favorable, over 90 per cent rooting was obtained in Cornus stolonifera, Lonicera tatarica, Rosa sp., Spiraea flexuosa, Syringa vulgaris, and Viburnum trilobum, while over 75 per cent rooting occurred in Corylus cornuta, Cotoneaster acutifolia, and Syringa villosa. combination of factors which resulted in maximum root production has been tabulated in the summary. The results obtained for many of the above species were poor, when the combination of influencing factors were unfavorable. In Corylus cornuta, and Cotoneaster acutifolia no cuttings rooted at 0 concentration, while Rosa sp., and Viburnum trilobum rooted poorly unless conditions were just right. Although the number of cuttings rooted, in those kinds which root with difficulty, e.g. Corylus and Cotoneaster, was very satisfactory and compared favorably with the easy sorts, the number of roots produced was considerably less than produced by the easily-rooted species. The greater the sensitivity of the plant to phytohormones the more roots there are produced.

Throughout the experiments it was found impossible to stimulate Amelanchier alnifolia to root production. In all, over

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1000 cuttings of this species were tested under a wide range of conditions and not a single root was produced.

The season when cuttings are taken from many plants is known to greatly influence the success of the rooting process. Unfortunately, this aspect of the rooting problem has not been as extensively studied for Alberta conditions as could be desired. Information on the influence of time of season to gather cuttings has been obtained for only a few species. However, the results on these few species demonstrate the importance of this factor in obtaining maximum results, and also the necessity of further testing of a large number of species. For most of the kinds studied, satisfactory rooting response was secured over a sufficient period of time to make possible the extensive propagation of these species by means of the greenwood cutting.

Rosa sp. was found to respond best when taken late in the season. Viburnum trilobum and Corylus cornuta root more satisfactorily when taken early in the season, about as soon as the new growth is sufficiently mature. Cornus stolonifera, Spiraea flexuosa and Syringa vulgaris are found to root very satisfactorily whether gathered on June 20 or July 15. The results of the 1938 investigations show quite clearly that all the above species, except Rosa sp., may be gathered from about June 15 to July 15, at least, and be expected to give close to maximum results. This period may vary somewhat with the climatic conditions and with further study may be extended towards the end of July.

The results proved quite conclusively that cuttings may be rooted satisfactorily under outdoor conditions, at least in north central Alberta. The results for such conditions were not only favorable, but in some cases they effected a greater response than

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that produced in the greenhouse at 70° F. The Rosa sp. seemed especially favorable to outdoor propagation. In fact, satisfactory rooting was obtained with it only in the outdoor frames. In Spiraea flexuosa and Cornus stolonifera the differences between the number of cuttings rooted at 70° F. and under outdoor conditions were negligible, but more roots were produced under the latter conditions. Thus, these three species may be propagated in a commercial way outdoors with just as desirable results as in the greenhouse. Although very satisfactory results were obtained in the outdoor frames, the rooted cuttings were never quite as fresh as those in the greenhouse frames, probably due to the lower humidity in the former case.

The three remaining species, Syringa vulgaris, Viburnum trilobum and Corylus cornuta responded best in the greenhouse at 70° F.

Although the 70° F. temperature was best for these species very
favorable results were secured outside. The evidence seems to be
that the lack of greenhouse facilities need not deter any nurseryman from propagating many of his own trees and shrubs rather than
buying lining-out stock. In most cases the uncontrolled (check)
temperature in the greenhouse was definitely the poorest.

SUMMARY AND CONCLUSIONS.

The rooting response of softwood cuttings of various species of plants was studied in relation to the influence of type of medium, temperature, phytohormones and concentrations of these, time of season at which the plant material is taken, and position of the basal cut. Special attention was given to the interrelationships of these variables, under Alberta conditions. From the results and analyses the following conclusions are drawn.

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- 1. Statistical analysis of the data reveals that strong interactions occur among many of the variables tested. The more important interactions are those involving temperature, species, treatments, concentrations and date of gathering the cuttings. This fact indicates that, in most cases, it is unsafe to recommend any one condition of a variable as being the best, without first stating the combination of other factors with which it is to be used.
- 2. As a guide for practical purposes, a summary table based upon the analyses and results is presented. This shows the particular combination of variables which, in these experiments, results in maximum rooting response. (see Tables VIII and XXII).
- 3. By the proper combination of phytohormones with the other factors, satisfactory rooting may be obtained in the following species of plants: Cornus stolonifera, Corylus cornuta, Cotoneaster acutifolia, Lonicera tatarica, Rosa sp., Spiraea flexuosa, Syringa villosa, Syringa vulgaris, and Viburnum trilobum.
- 4. Amelanchier alnifolia cannot be stimulated to root production by any of the earner methods used in these tests.
- 5. The order of response of the various species based upon the percentage of cuttings rooted, under optimum experimental conditions, is as follows; Cornus stolonifera, 100 per cent rooting; Lonicera tatarica, 97.5 per cent; Viburnum trilobum, 93.5 per cent; Rosa sp., 93.5 per cent; Syringa villosa, 87.5 per cent; Cotoneaster acutifolia, 85 per cent; and Corylus cornuta, 78.1 per cent.
- 6. The order of species for the number of roots produced, showing the average number of roots per cutting under optimum conditions,

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- is; Cornus stolonifera, 88.1 roots per cutting; Spiraea flexuosa, 76; Lonicera tatarica, 54; Syringa villosa, 41; Viburnum trilobum, 35.5; Rosa sp., 29; Syringa vulgaris, 21; Corylus cornuta, 10.5; and Cotoneaster acutifolia, 6.5;
- 7. The order of the various species in respect to the percentage of cuttings rooted is approximately the same as the order of the same species in respect to the number of roots produced per cutting.
- 8. The type of medium is a factor of minor importance in propagating greenwood cuttings, when phytohormones are used. Although for most species a sand medium gives slightly better results than a sand-and-peat mixture, in general this medium is not distinctly superior.
- 9. The results for outdoor conditions are very satisfactory. In terms of the whole experiment, the greatest response occurs in the outside frames, but the greenhouse at 70° F. is more often the better temperature when the remaining factors are in the most suitable combination. The outside condition is definitely better than greenhouse temperatures which are not thermostatically controlled. Where greenhouse facilities are not available, propagation of many species may be successfully carried on out-of-doors under Alberta conditions.
- 10. Temperature influences the stimulative action of the different phytohormones tested. Indole-acetic, indole-butyric and indole-propionic acids are more effective at 70° than at 60° F., whereas naphthalene-acetic acid produces greater stimulation at 60° F.
- 11. Temperature significantly affects the results with cuttings

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gathered at different times of the season. As the season advances the higher temperature (70° F.) is decidedly more advantageous than 60° F. Sixty degrees temperature is more effective early in the season.

- 12. The best concentration of phytohormone for each species studied is not the same for all sets of conditions within the experiment. The optimum concentration, along with the combination of variables, that produced the maximum rooting response is shown in Tables VIII and XXII.
- 13. Indole-butyric and indole-acetic acids are superior to indole-propionic and naphthalene-acetic acids in root stimulation, under the conditions of these investigations. Indole-butyric and indole-acetic acids are about equally effective, although indole-butyric acid may be slightly more stimulative in most cases.
- 14. The position of the basal cut is apparently a factor of very little importance, insofar as it influences the number of cuttings rooted. From the standpoint of the number of roots produced, it may have a slight effect. In only one case did the place of the basal cut sufficiently affect the number of roots produced to show a mathematical significance. Even in this instance, the differences exhibited are not outstanding so that this factor does not warrant more than minor consideration.
- 15. The best time to take greenwood cuttings of most species is comparatively early in the season. In Alberta, this is from about June 15 to July 15, or even extending a little later depending on seasonal conditions.
- 16. Cuttings of Corylus cornuta, and Viburnum trilobum respond

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best when taken early in the season, about June 20. The wood of these species should be taken as soon as it is sufficiently mature to handle as greenwood cuttings. Cornus stolonifera, Spiraea flexuosa, and Syringa vulgaris root best when taken about June 20, but produce more roots when the wood is more mature, that is, when taken about July 15. In all five species, the rooting response is very satisfactory whether taken June 20 or July 15, so that these species may be safely taken over that period of time.

17. Rosa sp. (probably R. acicularis) does not root satisfactorily when taken early in the season. Better results are obtained when the cuttings are gathered early in August, at which time the greenwood cuttings are well matured.

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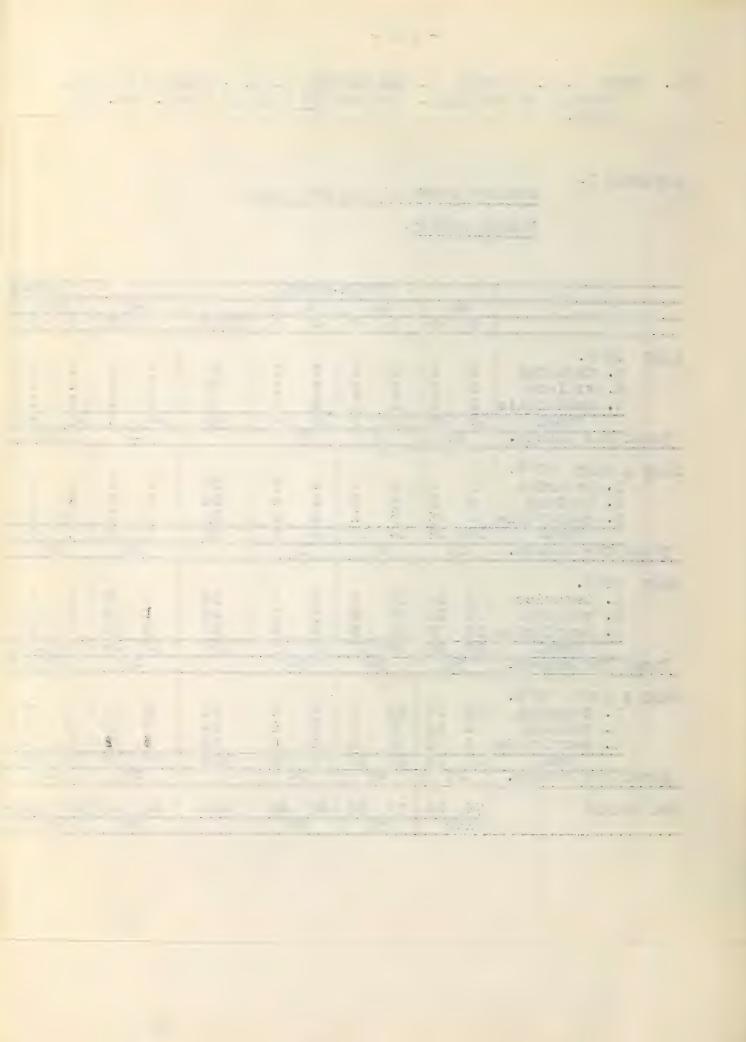
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APPENDIX I.

SUMMARY SHEET of the 1937 DATA.

NUMBER ROOTED.

		INDOLE	-ACETIC			NAPH-	ACETIC	1		INDOLE	-BUTYRIC			TN	DOT.R-	PROPION	TC	
	40	20	0		40	20	0		40	20	0		40		20	0	1	
	A B	A B	A B	TOTAL	A B	A B	A B	TOTAL	A B	A B	A B	TOTAL		BA	В	AE	TOTAL	TOTALS
SAND 60° F. L. tatarica S. villosa C. acutifolia Total Total for concen.	6 6 6 7 1 6 13 19	4 3 5 5 1 1 10 9	9 7 9 7 0 0 18 14	35 39 9 83	5 3 4 3 5 1 14 7	6 3 6 0 3 1 15 4	4 1 3 5 0 0 7 6	22 21 10 53	5 6 3 4 6 1 14 11	6 7 6 7 2 1 14 15	2 0 4 2 0 0 6 2	26 26 10 62	7 4 1	7 6 3 4 2 1 2 11	5 4 3 12 23	5 6 5 4 0 0	36 24 7 67	119 110 36 265
			<u> </u>									1				20		
SAND & PRAT 60°F. L. tatarica S. villosa C. acutifolia Total Total for concen.	3 4 4 3 6 5 13 12	6 4 5 8 3 1 14 13	6 5 2 4 0 0 8 9	28 26 15	3 3 1 1 3 1 7 5	6 5 1 3 0 1 7 9	1 3 6 4 0 0 7 7	21 16 5 42	7 4 5 8 2 4 14 16	9 9 9 8 0 1 18 18	1 1 4 6 0 0 5 7	31 40 7 78	6 3 1 10 1 24	9 4 3 6 2 0 4 10	3 6 1 10 20	5 2 7 7 0 0 12 9	29 32 4 65	109 114 31 254
SAND 70°F. L. tatarica S. villosa C. acutifolia Total Total for concen.	9 10 8 10 10 9 27 29	10 9	8 8 5 5 0 0 13 13	54 46 38 138	6 9 1 4 5 3 12 16	8 9 2 3 4 3 14 15	4 7 8 10 0 0 12 17 29	43 28 15 86	5 6 7 4 5 9 17 19	8 10 10 7 4 3 22 20 42	4 5 9 9 0 0 13 14 27	38 46 21 105	5 1 7 15	6 6 2 2 0 0 8 8 8	7 3 1 11	3 4 4 6 0 0 7 10	31 18 2 51	166 138 76 380
SAND & PRAT 70°F. L. tatarica S. villosa C. acutifolia Total Total for concen.	10 10 6 10 5 6 21 26	10 9 7 8 9 6 26 23	5 6 3 0 0 0 8 6	50 34 26 110	9 9 3 3 3 2 15 14	8 7 3 3 1 4 12 14	8 5 6 9 0 0 14 14 28	46 27 10 83	8 6 7 6 6 5 21 17	10 10 9 9 4 1 23 20 45	4 7 8 9 0 0 12 16 28	45 48 16 109	3 3 2 8 17	7 4 1 1 1 0 6	6 3 1 10	5 7 2 5 1 0 8 12	32 15 6 53	173 124 58 355
SUM TOTALS	74 86	79 72	47 42 89	400	48 42	48 42	40 44	264	66 63	77 73 150	36 39 75	354	37 43 80		43	37 41 78	236	1254



APPENDIX II.

SUMMARY SHEET of the 1937 DATA.

NUMBER of ROOTS.

			NAPH	-ACE	ric				IN	DOL	E-BUTYRIC		INDOLE-PROPIONIC												
	40		20	0		40		20		0			40	20		0			40	20		0			
	A	В	A B	A B	TOTAL	A.	В	A	В	A	В	TOTAL	A B	A	В	A B	TOTAL	A	В	A	В	A	В	TOTAL	TOTALS
SAND 60°F. L. tatarica S. villosa C. acutifolia Total Total for concen.	235 2 3	28 81 9	85 142 327 270 2 3 414 415 829	56 55 87 95 	586 1295 17 1898	13	89 110 2 201	96 165 5 266 294		28 75 - 103 158	1 54 - 55	315 435 22 772	156 159 312 184 17 1 485 344	133 2	88 35 3	3 0 25 33 28 33 61	728 922 26 1676	8 1	6 20 1 3	86 43 2 131 251	60 55 5 120		68 56 - 24	377 235 11 623	2006 2887 76 4969
SAND & PRAT 60° F. L. tatarica S. villosa C. acutifolia Total Total for concen.		87 87 12 86	211 116 118 291 5 3 334 410 744	50 82 27 28 77 110 187	603 606 41 1250	42 15 17 74	48 3 2 53	-	71 150 2 223	-	11 67 78	276 297 21 594	283 109 121 217 5 20 409 346 755	227 2	35 16 3 54	1 1 42 79 	1049 902 28 1979			-	39 148 1 188	15 70 10 - 85 10 194	-	364 526 7 897	2292 2331 97 4720
SAND 70° F. L. tatarica S. villosa C. acutifolia Total Total for concen.	346 4	56 40 61 57	488 262 314 237 60 56 862 555 1417	19 32 67 71 86 103 189	1525 1475 250 3250	18 19	246 51 34 331	218 16 23 257 433	-	12 95 1 - 107 1 296	41 - 89	757 398 86 1241	138 183 293 240 47 77 478 500 978	530 2 15		41 56 91 90 132 146 278	1498 1485 151 3134	12	1 31 1 -	-	152 59 1 212	7 46 11 -53 14 198	33 12 - 45	654 284 2 940	4434 3642 489 8565
SAND & PEAT 70° F. L. tatarica S. villosa C. acutifolia Total Total for concen.	221 4	37 306	283 161 157 254 65 37 505 452 957	22 41 22 0 44 41 85	1088 1099 176 2363	16	242 36 11 289	60	100 75 17 192	23 50 1 - 73 1 229	56	765 403 49 1217	354 288 300 343 56 57 710 688 1398	406 2 29	71 67 7 45	12 28 78 82 	1642 1476 149 3267	100	3 12 4 2	105 15 3 123 20			11 05 -	435 179 12 626	3930 3157 386 7473
SUM TOTALS	1693 23		2115 18 3 2 3947	350 404 754	8761	732	874	789 140		332 4 810		3824	2082 1878 3960	2762 26 5434	72	293 369 662	10056		571 1071	658 126		240 51 754	4	3086	25727

SUMMARY SHEET of the 1938 DATA. NUMBER ROOTED.

CDECTES	GIRL TOWN THE GARLE		TOTAL MOTTON		DOCA CD
SPECIES	SYRINGA VULGARIS		JRNUM TRILOBUM	CORNUS STOLONIFERA	ROSA SP. 40 20 0
HORMODIN A CONCENTRATION PLACE OF BASAL CUT	40 20 0 A B A B A B TOTAL	A B A	B A B TOTAL	40 20 0 A B A B A B TOTAL	A B A B A B TOTAL
SAND 60° F Date 1	7 7 7 8 3 4 36	5 5 6			
n 2	3 2 3 4 2 0 14 2 0 3 2 1 3 11	0 0 5	8 7 8 39 3 3 5 16 2 2 4 13	7 8 7 8 6 8 44	$\begin{bmatrix} 2 & 4 & 4 & 5 & 0 & 0 & 15 \\ 0 & 0 & 3 & 5 & 0 & 1 & 9 \\ 1 & 0 & 4 & 0 & 0 & 2 & 7 \end{bmatrix}$
Total	12 9 13 14 6 7 61	7 5 14	13 12 17 68	20 22 23 23 14 16 118	3 4 11 10 0 3 31
Total for Concen.	21 27 13	12 2	27 29	42 46 30	7 21 3
SAND & PEAT 60° F Date 1	8 8 8 8 4 5 41 6 7 3 2 1 1 20	5 6 8	8 7 8 42	8 8 6 8 6 7 43 7 7 8 8 8 4 42	1 2 6 5 1 0 15 0 1 0 1 0 0 2
# 2 # 3	1 1 1 0 0 0 3	0 0 3 2 0 3	8 7 8 42 2 4 4 13 5 3 5 18	7 5 8 7 0 0 27	1 2 7 4 5 0 19
Total for Concen.	15 16 12 10 5 6 64 31 22 11	7 6 14 13 2	15 14 17 73 29 31	22 20 22 23 14 11 112 42 45 25	2 5 13 10 6 0 36 7 23 6
SAND 70°F Date 1 2	7 7 7 8 4 0 33 8 8 8 7 47	7 6 7	6 7 7 40 7 8 7 29	8 8 8 8 8 8 48 8 8 8 7 8 7 46	0 0 1 3 0 0 4 0 1 2 3 0 0 6
" 3 Total	8 8 8 8 8 7 47 6 6 5 5 3 2 27 21 21 20 21 15 9 107	3 4 3 10 11 16	7 8 7 29 7 7 5 29 20 22 19 98	5 7 8 4 1 2 27 21 23 24 19 17 17 121	1 4 7 5 1 2 20 1 5 10 11 1 2 30
Total for Concen.	42 41 24		36 41	44 43 34	6 21 3
SAND & PEAT 70°F Date 1	8 5 7 6 5 2 33	7 A (9 4 5 70	4 0 0 0 E 0 41	0 0 1 0 0 0 1
1 2 11 3	8 5 7 6 5 2 33 7 8 6 7 5 3 36 1 0 1 0 0 0 2	3 4 6 0 0 5 3 1 3	8 4 7 32 3 7 6 21 4 6 3 20	6 7 8 8 5 7 41 8 8 8 7 5 8 44 7 6 7 4 0 0 24	0 1 4 3 2 0 10 2 1 3 5 1 0 12
Total	16 13 14 13 10 5 71	6 5 14	15 17 16 73	21 21 23 19 10 15 109	2 2 8 8 3 0 23
Total for Concen.	29 27 15	11 2	29 33	42 42 25	4 16 3
SAND Out. Date 1	6 8 5 3 3 5 30	6 7 6	8 8 8 43 2 7 6 20	8 7 8 8 8 8 47	8 7 5 4 2 3 29
n 2	6 8 5 3 3 5 30 6 6 7 6 6 0 31 0 0 2 1 0 0 3	0 0 5	2 7 6 20 6 4 4 18	8 7 8 8 8 8 47 8 8 7 8 6 6 43 5 3 6 6 0 0 20	8 7 5 4 2 3 29 3 3 4 7 4 24 6 4 7 8 3 7 35
Total for Concen.	12 14 14 10 9 5 64 26 24 14	6 7 15	16 19 18 81 37	21 18 21 22 14 14 110 - 39 43 28	17 14 15 16 12 14 88 31 31 26
SAND &	20 24 14	19 9) 1	77 47 20	
PRAT Out. Date 1	7 8 8 7 5 5 40 8 7 7 6 7 2 37 1 6 4 4 0 1 10	3 6 6	8 6 7 36 4 4 5 17 6 5 4 22	8 8 8 8 5 8 45 8 8 7 8 7 8 46 5 3 3 5 1 0 17	5 4 5 7 3 2 26 4 4 5 4 8 5 30 7 7 8 7 2 3 34
* 3		0 1 3 6			7 7 8 7 2 3 34 16 15 18 18 13 10 90
Total for Concen.	16 15 19 17 12 8 87 31 36 20	3 8 15 11 3	18 15 16 75 33 31	21 19 18 21 13 16 108 40 39 29	31 36 23
SUM TOTALS Date 1 2	43 43 42 40 24 21 213	29 34 39 0 2 27	46 39 45 232 21 33 33 116	46 46 46 48 40 45 271 46 47 45 46 40 41 265	16 17 22 24 6 5 90 7 10 17 20 17 10 81 18 18 36 29 12 14 127
# 2 # 3	43 43 42 40 24 21 213 38 38 34 33 29 13 185 11 7 16 12 4 6 56	10 6 22	30 27 25 120		18 18 36 29 12 14 127
	92 88 92 85 57 40 454	39 42 88	97 99 103 468	126 123 131 127 82 89 678	41 45 75 73 35 29 298

			ROSA	SP						CORY	LUS	CORI	WTA				SPI	REA	FLE	XUOS	A	
	40		20			0		40)	20)	C)		40)	20			0		
L	A	В	A	В	A	В	TOTAL	A	В	A	ㅂ	A	В	TOTAL	A	В	A	В	A	В	TOTAL	TOTALS
	2 0 1	4 0 0	4 3 4	5 5 0	0 0	0 1 2	15 9 7	6 1 0	4 0 1	3 0 1	4 0 1	0 0 0	1 0 0	18 1 3	7 7 0	8 7 0	8 6 1	8 6 0	3 0 0	2 0 0	36 26 1	191 110 62
3	3	4		10	0	3	31	7	5	4	5	0	1	22	14	15		14	3	2	63	363
	7		21			3		12	-		<u> </u>	1			29		29			5		
	1 0 1 2	2 1 2 5	6 0 7 13	5 1 4 10	1 0 5	0 0 0 0	15 2 19 36	5 1 0 6	8 1 0	0 1 4	2 0 0 2	0 0 0	0 0 0	15 3 4 22	8 6 0 14	8 4 0	7 5 1 13	7 5 3	0 1 0	2 2 0 4	32 23 4 59	188 103 75 366
3	0 0 1	0 1 4 5	1 2 7 10	3 3 5	0 0 1 1	0 0 2 2	4 6 20 30	7 3 0	6 2 0 8	531	2 3 3 8	0 0 0	0 0 0	20 11 4 35	8 5 0	7 6 4 17	8 8 1	7 7 2 16	3 2 1 6	3 4 0	36 32 8 76	181 171 115 467
al orb orb	0 0 2	0 1 1	1 4 3	0 3 5	0 2 1	0 0 0	1 10 12	5 4 0	7 2 1	1, 6, 6	2 3 3	0 0	0 0 0	15 15 10	8 6 0	8 4 0	8 6 2	7 5 2	2 1 0	0 3 0	33 25 4	155 151 72
7	2	2	8	8	3	0	23	9	10	13	8	0	0	40	14	12		14	3	3	62	378
	4	1	16	<u> </u>		3		19)	2]	L	0			26		30		(6		
7 5 0	8 3 6	7 3 4	537	4 4 8	2 7 3	3 4 7	29 24 35	4 0 1	5 3 0	3 0 1	5 1 1	0 0	0 0	17 4 3	5 8 1	6 8 4	7 7 7	8 7 3	2 2 0	5 2 1	33 34 16	199 156 95
0	17	14	15	16	12	14	88	5	8	4	7	0	0	24		18		18	4	8	83	450
567	5 4 7 16 3:	4 4 7 15	5 5 8 18	7 4 7 18	3 8 2 13	2 5 3	26 30 34 90	2 0 4 6	3 2 0 5	1 0 2 3	0 2 5	0 0 0	0 0 0	6 4 11 21	8 7 2 17	7 7 4 18	39 4 6 5 15	8 7 3	2 4 0 6	2 2 1 5	31 33 15 79	184 167 109 460
5 2 8	16 7 18 41	17 10 18	22 17 36 75	24 20 29	6 17 12 35	5 10 14 29	90 81 127 298	29 9 5 43	33 10 2 45	13 10 15 38	15 9 13 37	0 0 0	1 0 0	91 38 35 164	44 39 3	44 36 12 92	17 :	37 13		14 13 2 29	201 173 48 422	1098 858 528 2484

SUMMARY SHEET of the 1938 DATA.

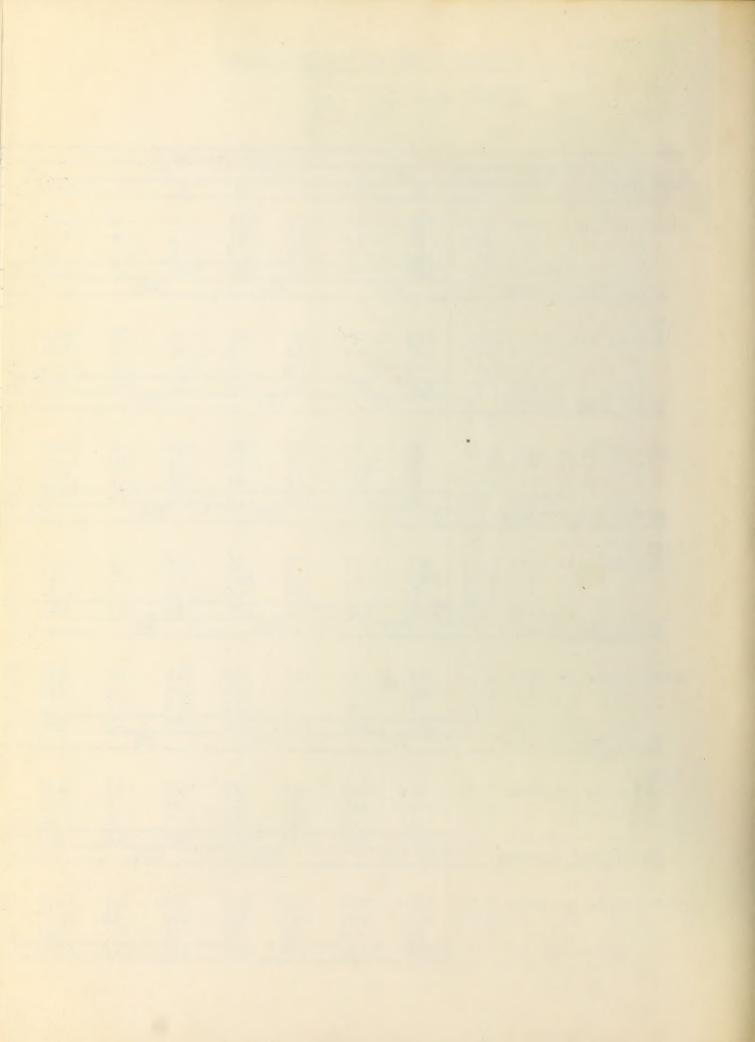
NUMBER of ROOTS.

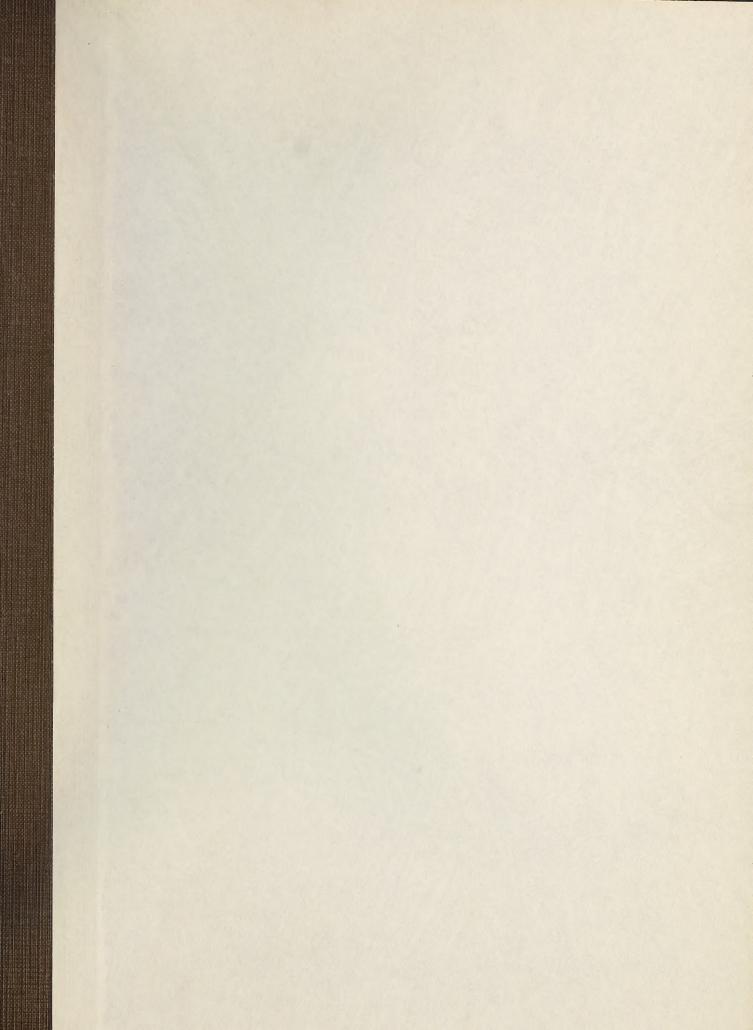
SPECIES	SYRINGA VULGARIS N 40 20 0										NUM TR	ILOBUM					CORNU		LONIFE	ERA		L		R
HORMODIN A CONCENTRATION PLACE of BASAL CUT	40 A	В	<u>A</u>	В	A	В	TOTAL	A 4	-0 B	<u>A</u>	0 B	0 A	В	TOTAL	A	10 B	20 A	В	A	В	TOTAL	A 40	0 B	20 A
SAND 60°F Date 1 " 2 " 3 Total Total for Concen.	81 31 15 127 21	28 56 0 84	37 18 12 67	34 55 4 93	8 5 2 15	6 0 7 13	194 165 40 399	140 0 8 148	138 0 0 138	142 187 10 339	251 39 70 360	89 34 19 142 49	192 102 55 349	952 362 162 1476	838	275 611 99	303 591	326 515 199 1040	159 107 0 266	126 164 39 329	1482	27 0 1 28	74 0 10 84	84 19 108 211 410
SAND & PEAT 60 F Date 1 " 2 " 3 Total Total for Concen.	65 83 6 154 28	68 60 3 131	33 14 5 52	25 5 0 30 2	9 6 0 15	10 8 0 18	210 176 14 400	97 0 15 112	223 0 0 223	379 126 130 635	292 125 96 513	54 62 13 129	155 118 134 407	1200 431 388 2019	243 366 156 765	350 589 31 970	125 485 123 733 157	199 527 117 843	98 168 0 266	86 62 0 148	427	9 0 7 16	10 18 7 35	82 0 125 207 304
SAND 70°F Date 1 " 2 " 3 Total Total for Concen.	68 235 48 351 58	60 121 51 232	38 77 32 147 30	27 88 41 156	23 36 13 . 72	0 34 6 40	216 591 191 998		226 38 118 382	251 270 211 732	328 257 385 970	128 162 184 474	202 211 257 670	1407 938 1236 3581	1225	425 616 255 1296 521	248 420 270 938 200		221 172 1 394	234 101 15 350 44	1741 2536 991 5268	0 0 15 15	0 61 51 112	15 24 80 119 2 405
SAND & PEAT 70°F Date 1 " 2 " 3 Total Total for Concen.	68 169 8 245 407	35 127 0 162	37 60 14 111 25	18 123 0 141	19 31 0 50	5 9 0 14	182 519 22 723	104 0 32 136	101 0 12 113	140 206 43 389	224 161 147 532 21	49 96 170 315	180 187 26 393	798 650 430 1878	132 650 197 979	245 775 356 1376 55		265 517 164 946	23 33 0 56	74 104 0 178 34	2512 1028	0 0 45 45	0 15 16 31	7 16 92 115 229
SAND Out Date 1 " 2 " 3 Total Total for Concen.	44 66 0 110 278	59 109 0 168	23 96 5 124	16 80 10 106	11 24 0 35	17 0 0 17	170 375 15 560		180 0 0 180	123 231 51 405	236 115 46 397 02	252 146 83 481	287 136 67 490	1206 628 247 2081	69	221 826 35 1082	174	360 559 121 1040	122 145 0 267	149 140 0 289	1599 2776 399 4774	175 116 236 527	93 156 119 368	15 106 95 216 515
SAND & PEAT Out Date 1 " 2 " 3 Total Total for Concen.	61 104 1 166 400		55 64 13 1132 24	28 73 15 116	23 44 0 67	17 5 1 23	297 411 30 738	69 0 0 69	264 5 4 273	175 105 138 418	225 92 73 390 08	59 39 61 159	158 64 68 290	950 305 344 1599	743		349 426 33 808 1650	842	81 154 3 238	124 171 0 295	1611 2542 409 4562	95 101 116 312 66	78 43 227 348	90 39 247 1 376 629
SUM TOTALS Date 1	387 688 78 1153	363 594 54 1011	223 329 81 633	148 424 70 642	93 146 15 254	55 56 14 125	1269 2237 312 3818	810 0 136 946	1132 43 134 1309	1210 1125 583 2918	1556 789 817 3162	530	1174 818 607 2599	6513 3314 2807 12634	787	936	1721 1 2903 3 1025 1 5649 5	L020	704 779 4 1487	793 742 54 1589	8560 14975 3826 27361	306 217 420 943	255 293 430 978	293 204 3 747 1244 12

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40 A	В	A 20	В	O	В	TOTAL	40 A	В	20 A	В	0 	В	TOTAL	A	10 B	A	20 B	O	В	TOTAL	TOTALS
27 0 1 28	74 0 10 84	84 19 108 211	63 91 45	0 0 0	0 2 5 7	248 112 169 529	65 6 0 71 12	42 0 7 49	18 0 20 38	68 0 20 88	0 0 0	5 0 0 5	198 6 47 251	332 541 15 888	399 489 0 888	421 388 3 812	460 374 1 835	68 0 0 68	70 0 0 70 138	1750 1792 19 3561	4824 4849 1009 10682
9 0 7 16	10 18 7 35	82 0 125 207	52 5 40 97	2 0 13 15	0 0 0 0	155 23 192 370	22 11 0 33	80 1 0 81 4	0 2 17 19	8 0 0 8	0 0 0	0 0 0 0	110 14 17 141	378 376 0 754	162 328 0 490	174 193 43 410	244 304 133 681	0 30 0 30	33 43 0 76	991 1274 176 2441	3767 4115 1214 9096
0 0 15 15	0 61 51 112	15 24 80 119 40	125 109 52 286	0 0 2 2	0 0 9 9	140 194 209 543	78 14 0 92	77 6 0 83	59 11 4 74	3 9 15 27	0 0 0	0 0 0 0	217 40 19 276	455 290 0 745	216 401 127 744	293 368 3 664	146 277 7 430	26 10 8 44	21 55 0 76	1157 1401 145 2703	4878 5700 2791 13369
0 0 45 45	0 15 16 31	7 16 92 115	0 12 102 114	0 3 1 4	0 0 0 0	7 46 256 309	24 30 0 54	85 9 5 99	7 20 26 53	109 17 33 159	0 0 0	0 0 0	225 76 64 365	274 319 0 593	218 171 0 389	175 152 65 392	269 116 66 451	76 7 0 83	0 33 0 33	1012 798 131 1941	3250 4601 1931 9782
175 116 236 527	93 156 119 368	15 106 95 216	27 77 195 299	7 30 12 49	6 12 19 37	323 497 676 1496	63 0 11 74	88 93 0 181	56 0 7 63	81 3 4 88	0 0 0	0 0 0 0	288 96 22 406	320 810 6 1136 22	350 538 202 1090	361 612 502 1475	470 340 102 912	35 18 0	98 36 0 134	1634 2354 812 4800	5220 6726 2171 14117
95 101 116 312	78 43 227 348	90 39 247 376	77 48 128 253	9 28 3 40	5 12 4	354 271 725 1350	48 0 94	15 8 0	12 0 26 38	0 8 22 30	0 0 0	0 0 0 0	75 16 142 233	524 462	306 472 116 894	198 218 142 558	487 218 49 754	16 74 0	77 15 1	1608 1459 405 3472	4895 5004 2055 11954
306 217 420 943	255 293 430 978	293 204 747 1244	344 342 562 1248	18 61 31 110		1227 1143 2227 4597	300 61 105 466	387 117 12 516	152 33 100 285	269 37 94 400	0 0 0	5 0 0	1113 248 311 1672	2283 2798 118	1651 2399 445 4495		2076 1629 358 4063	221 139 368	299 182 1	8152 9078 1688 18918	26834 30995 11171 69000

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